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A NEW SPECIES OF MORTIERELLA

C. P. SIDERIS AND G. E. PAXTON

(WITH PLATE 12)

During the isolation of various parasitic as well as saprophytic fungi from diseased roots of pineapple plants a Phycomycete was obtained that refused to sporulate for a very long time. This organism finally sporulated while growing on a corn-meal agar media prepared in our laboratory.

The corn-meal decoction was obtained by partial hydrolysis of 50 grams of corn-meal with 10 cc. 1/*N* HCl in 500 cc. of water for 2 hours, at 80° C. The acid was neutralized at the end of this period by a corresponding volume of 1/*N* NaOH, and then 1 gram of trypsin, obtained from the Digestive Ferments Co., was added for further hydrolysis. The mixture was allowed to stand for 48 hours at 40° C. At the end of this period the fluid portion was removed by filtration through cotton and qualitative filter paper and then made to 2,000 cc. with additional tap water. The amount of agar agar added was 2 per cent.

This organism, as well as others that we have in our laboratory, has been found to sporulate more readily on these media than on any of the other standard media used so far in these studies. It has produced sporangia with perfect spores but no zygospores. Sporangia, but without perfect spores, were also produced by the same organism in a number of other media.

The organism differs considerably from various other *Mortierella* species in that its sporangiophores are considerably smaller than those of any of the other known species (FIG. 1). The spo-

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rangiophores emerge from non-differentiated hyphae and stand always above the media. They may or may not have rhizoids. Gemmae have also been observed very often on malt peptone agar, but not as often on corn-meal agar.

The sporangial membrane is very fragile, which fact may account for the rare occurrence of non-bursted ripe sporangia. The slightest amount of friction is capable of causing the mature sporangia to burst and set their spores free in the surrounding media. The spores vary in shape from spherical to ovoid and measure in width from 2 to 3 μ and in length from 4 to 7 μ. The mycelium varies considerably in thickness, measuring on the average about 6 μ, but in extreme cases it has been found as thick as 10 μ and as thin as 1 μ.

Mortierella elasson nov. sp.

Sporangiophores long, from 200 to 500 μ, wide at the base, from 5 to 10 μ, and at the tip from 3 to 6 μ, not branched, non-septate, colorless and having from none to few rhizoids. Sporangia from 10 to 24 μ in diameter, spherical or slightly ellipsoid, colorless, varying considerably in the number of their spores and very fragile during maturity. Spores commonly oval, but sometimes spherical or tetrakaidecahedric, 3 to 6 μ wide and 5 to 10 μ long and in rare cases few may be twice or three times as large. Zygospores never observed. It grows saprophytically on dead pineapple roots. It was obtained from roots of pineapples grown on the islands of Oahu and Maui, Territory of Hawaii.

The specific name *elasson* of the organism is taken from the Greek word ἐλάσσων = minor, owing to the small size of its sporangiophores.

A comparative study of this and other species indicates that the size of the sporangiophores, sporangia and spores is considerably smaller than that of any of the species described so far. The species *M. simplex* Van Tieghem & Le Monnier, *M. Rostafinskii* Brefeld, and *M. strangulata* Van Tieghem with which this organism is more closely related than any others have sporangiophores measuring in length between 600 and 1,000 μ, in width at the base between 50 and 100 μ and at the tip between 15 and 30 μ. The sporangia in these same species measure between 50 and 120 μ in diameter, and the sporangiospores

between 5 and 10 μ. There cannot be any doubt whatsoever but that this organism constitutes a new species.

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EXPLANATION OF PLATE 12

Fig. 1. Colony on corn-meal agar 10 days old; 2. Hyphae. (× 150); 3. Sporangium and spores. (× 300); 4. Spores. (× 600); 5. Sporangiophore with sporangium intact. (× 300); 6. Sporangiophore with collapsed sporangium. (× 300); 7. Sporangia. (× 600); 8. Spores. (× 300); 9. Germinating spore. (× 600); 10. Hyphae. (× 300.)

STUDIES IN TROPICAL ASCOMYCETES—VI. PHYLLACHORA SIMABAE CEDRONIS

FRED J. SEAVER

(WITH 2 TEXT FIGURES)

In the fifth installment of this series of articles the writer published a supposedly new species of *Phyllachora* from South America on an unnamed host. Shortly after this article appeared

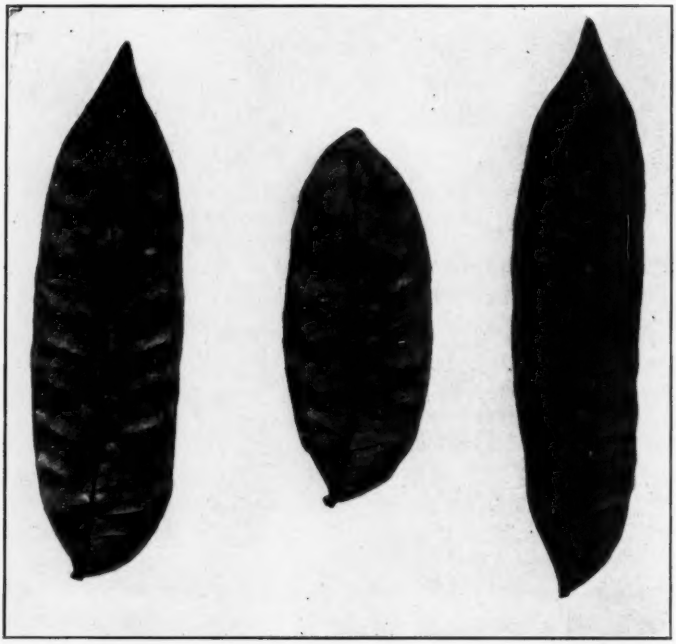


FIG. 1. *Phyllachora Simabae Cedronis* on small leaves on *Simaba Cedron* Pl. from Costa Rica. Taken from the phanerogamic collection of The New York Botanical Garden. A. Tonduz *9948*.

a letter was received from Dr. H. Sydow of Germany, stating that our *Phyllachora Pennellii* was the same as the Costa Rican *Phyllachora Simabae Cedronis* P. Henn.

Following this suggestion the writer went through the phanerogamic collection of The New York Botanical Garden looking over herbarium specimens of this host. As a result of this search a specimen of *Phyllachora* resembling *Phyllachora Pennellii* was obtained on leaves collected in Costa Rica. This specimen although resembling our species differed slightly in general appearance and the writer was still unconvinced that *Phyllachora Pennellii* and *Phyllachora Simabae Cedronis* were the same thing.

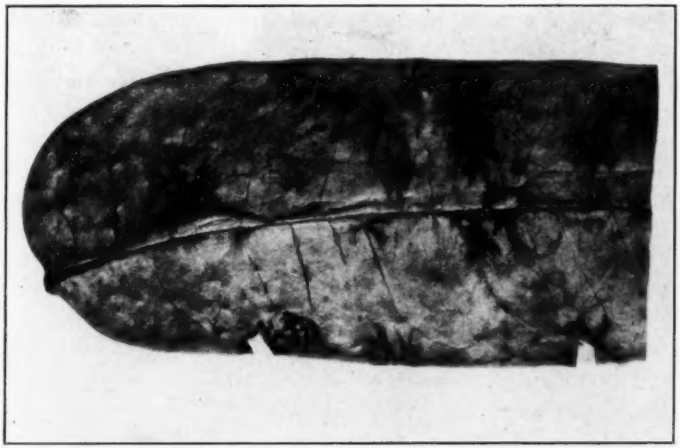


FIG. 2. Photograph of the type of *Phyllachora Simabae Cedronis* loaned by H. Sydow from Germany. Cf. Mycologia **20**: *pl. 26, f. 1.*

At the request of the writer, Dr. H. Sydow later loaned to us the type specimen of P. Hennings' species, a photograph of which is here reproduced. This proves conclusively that the two are identical. Had we been able to name the host on which our species occurred this oversight would doubtless not have occurred but inasmuch as Dr. Pennell did not know the host it was impossible to use this as a clue in running down the species. As often happens the fungus in this case has enabled us to identify the host. The synonymy of the species would then be as follows:

PHYLLACHORA SIMABAE CEDRONIS P. Henn. Hedwigia **43**: 147. 1904.

Phyllachora Pennellii Seaver, Mycologia **20**: 222. 1928.

THE NEW YORK BOTANICAL GARDEN

NOTES ON THE PARASITIC FUNGI OF ILLINOIS—IV

L. R. Tehon and G. L. Stout

(With Plate 13)

Specimens obtained while prosecuting the plant disease survey of Illinois, which has been under way as one of the activities of the Illinois State Natural History Survey since 1922, continue to supply new and interesting examples of parasitic fungi. This paper, in common with our previous "Notes," is devoted chiefly to the description of new non-economic forms; but we have appended an additional list of species, either not hitherto known to occur in Illinois or with new localities and notes on distribution.

Type specimens upon which our novelties are based are denoted by their accession numbers in the Mycological Collection of the Natural History Survey. When the type material is abundant, a deposit is being made also with the New York Botanical Garden. The names of wild hosts are those given in the 7th edition of Gray's Manual.

We are constrained, in our treatment of the Ascomycetes and pycnidial forms, to make a limited use of the newer European classifications, for which the discussion of the individual cases will show the convenience.

Stigmatophragmia Tehon & Stout, n. gen.

Genus of the Hemisphaeriales, family Stigmateaceae. Perithecia subcuticular, hemispheric, membranous to carbonous. Perithecial cover pseudoparenchymatic. Paraphyses present. Ascospores hyaline, oblong to fusoid, several septate.

This genus, based on the species subsequently described, is very certainly allied with the Hemisphaeriales. Most of its characters are those of the Hemisphaeriaceae; but its perithecia, which lie within the host, appear to have developed beneath the cuticular membrane, and this character alone, to which much weight is given by Sydow, throws it definitely out of this family

and unites it at once with the Stigmateaceae, while the pseudoparenchymatic perithecial cover, which very evidently develops from a truly radiate beginning, makes it distinctive among the genera of this family. It appears to parallel *Stigmatodothis* in most respects but has true paraphyses. Its position may be outlined as follows:

Perithecial membrane distinctly radiate...*Stigmatea*, *Stigmatodothis*, etc.
Perithecial membrane pseudoparenchymatic.
  Paraphyses present.
    Spore 2-celled, brown; ascoma setate.....*Coleroa*
    Spore several-celled, hyaline; ascoma smooth.....*Stigmatophragmia*
  Paraphyses absent.....*Aphysa*

Stigmatophragmia sassafrasicola Tehon & Stout, n. sp.

Foliicolous. Spots diaphyllous, circular or somewhat angular, 3–5 mm. in diameter, tan to brown epiphyllously, never cinereous, not friable, definitely limited by a distinct, very fine, unraised, purplish margin; characters similar hypophyllously but obscured by leaf bloom. Thyrothecia few, widely scattered, hypophyllous only, round, 200–225 μ in diameter, subcuticular, opening by a somewhat umbonate, carbonaceous, round ostiole 18–34 μ wide; thyrothecial membrane pseudoparenchymatic, non-radiate. Paraphyses filiform, unbranched, abundant, equalling or slightly exceeding the height of the asci. Asci cylindric, thin walled, abruptly rounded at the apex and the base, short-stiped, 8-spored, 65–80 $\times$ 10–15 μ. Spores hyaline, 3-septate, cylindric with tapered end-cells, or spindle-shaped, straight or slightly flexed, outline constricted at the septa, 14–17 $\times$ 3–4 μ; individual cells of equal length. PLATE 13, FIG. 1.

On *Sassafras variifolium*.

Seymour, Champaign County, October 15, 1925. Acc. No. *20103* (type).

On the same leaves are spots bearing the acervuli of the very common *Gloeosporium affinis;* but there seems to be no connection between the two fungi. This and the type of the new genus *Pseudodictya* described on a later page were taken from the same tree.

Melanospora interna Tehon & Stout, n. sp.

Perithecia developed in the pith cavity but not imbedded in the pith, very abundant, scattered, discrete, with a few hyaline mycelial hairs, translucent, golden-yellow, globose, rostrate,

150–300 μ in diameter; cavity 125–270 μ in diameter; rostrum 50–135 μ high, cylindrical, 40–55 μ wide, without an apical fringe of hairs. Asci saccate to very broadly clavate, evanescent, disappearing as the spores mature, 40–55 × 17–21 μ. Spores 8 per ascus, biseriate as a rule, broadly spindle-shaped, continuous, chocolate brown but with a small, light, almost hyaline region at each end, whence come the germ-tubes, 19.5–22 μ long, 8.5–11 μ broad, the walls marked with coarse, irregular reticulations. PLATE 13, FIG. 2.

On *Lycopersicon esculentum*.

Mound City, Pulaski County, November 13, 1927. Acc. No. *20939* (type).

This form is amply distinct from the species hitherto described. Between it and *M. Solani* Zukal there are few points of agreement, for the larger perithecia, the very long and comparatively wide rostrum, the narrower and longer asci, and the very much larger spores separate it completely. The habit, also, may be significant, as the plant from which it was taken appeared to be suffering from a type of root rot, the cause of which may have been this fungus, whose perithecia lined the cavity in the lower stem where the pith had disappeared.

Taxonomic characters within this genus are more striking than is usual in distinctive groups. The brilliant yellow perithecium, the usually very evident rostrum, the remarkable shape of the spores, and the typically evanescent asci serve to ally the forms now known. Among the species, *M. carpophila* Zukal is segregated by the possession of paraphyses; *M. antarctica* Speg. and *M. Marchaliana* Bomm. are at once allied to each other and separated from others by their long, cylindrical asci, in which the spores are disposed uniseriately; *M. ornata* Zukal and our *M. interna* possess spores with reticulate walls; and the remaining species, over 30 in number, though closely similar, appear readily distinguishable by means of such characters as spore-size, ascus measurements, and the like.

Metasphaeria Asparagi Tehon & Stout, n. sp.

Caulicolous. Maculicole rather than cankerous, the spots taking the form of much elongated, rather wide, gray lesions, from which the loosened cuticle falls away and exposes the perithecia, which are seated on the woody tissue beneath. Perithecia

numerous, scattered, subglobose, membranous, dark brown or carbonaceous, 220–375 μ in diameter, opening by means of a papillate, usually carbonized ostiole 6–14 μ in diameter. Asci long-clavate, verging to cylindrical, straight or (when from the sides of the perithecia) curved, short-stalked, double-walled, 75–130 × 11–16 μ. The inner wall of the ascus, with its content of spores, often slips out of the external sheath. Paraphyses hyaline, filamentous, 1–1.5 μ wide, equaling or exceeding the height of the asci. Ascospores hyaline, 3–5- but usually 4-septate, oblong, 17–25 × 5–6.5 μ, constricted at the septa, the second cell from the top (and sometimes adjacent cells) nearly spherical. PLATE 13, FIG. 3.

On *Asparagus officinalis*.

Anna, Union County, November 11, 1926. Acc. No. *19944* (type).

With this fungus there is found in some lesions a *Phoma*, recorded on another page as *P. asparagina*, which may be a pycnidial form—at least, there is no evidence of distinct mycelia.

Metasphaeria sassafrasicola Tehon & Stout, n. sp.

Foliicolous. Spots diaphyllous, subcircular, tan, with a narrow, dark-brown border, 3–7 mm. in diameter, occasionally confluent. Perithecia scattered, not gregarious, membranous, developed in and occupying the mesophyll, spherical, 75–100 μ in diameter; ostiole erumpent epiphyllously, papillate, somewhat carbonized, its opening 15 to 20 μ in diameter. Asci few, 6–10 per perithecium, oblong, with a short, blunt foot, uniformly 44–45 × 12–13 μ. Paraphyses few, filiform, equaling the asci in height. Ascospores 8 per ascus, 3-septate, hyaline, arranged either irregularly or in 2 bundles of 4 each, 16–18 × 2.2–2.4 μ; the pre-apical cell round. PLATE 13, FIG. 6.

On *Sassafras variifolium*.

Seymour, Champaign County, October 15, 1925. Acc. No. *20103* (type).

Pleospora Oleraceae Tehon & Stout, n. sp.

Foliicolous. Spots chiefly circular, sometimes oval, 0.75–5 mm. in diameter, white, with a distinctly raised border, the interior collapsed, papery, and translucent. Perithecia innate, membranous, spherical, 65–100 μ in diameter, erumpent epiphyllously. Ostiole circular, only slightly raised, 20–40 μ in diameter. Paraphyses filiform, equaling the asci. Asci few,

thin-walled, short-stalked, asymmetrically oval, 45–48 × 18–21 μ. Spores crowded, 8 per ascus, smoky to distinctly olivaceous, oval, thick-walled, with 3 or 4 horizontal septa, medial cells variously divided longitudinally, 22–28 × 8.8–11.0 μ, never more than very slightly constricted. PLATE 13, FIG. 4.

On *Brassica oleracea*.

West Vienna, Johnson County, July 7, 1926. Acc. No. *19358* (type).

This is quite distinct from *P. herbarum* var. *Brassicae* (Lasch) Sacc., in which the perithecia are three or more times as wide, the asci nearly three times as long, and the spores half again as wide. It falls in Saccardo's section Eu-Pleospora.

Phyllosticta Rugelii Tehon & Stout, n. sp.

Foliicolous. Spots diaphyllous, irregularly circular, dark-brown and concolorous above and below, faintly marked concentrically by the collapse of diseased tissue, 2–10 mm. in diameter. Pycnidia borne in circular to oval, unbordered, cinereous, deciduous areas 1–3 mm. in diameter, abundant, scattered, wholly membranous, translucent, spherical to slightly applanate, 35–65 μ in diameter, developed in and occupying the collapsed mesophyll, opening only through the epiphyll by means of a long-papillate ostiole 4–8 μ wide. Spores hyaline, continuous, chiefly elliptic but with oval and asymmetrically allantoid variants, 6.5–8.5 × 2–3 μ.

On *Plantago Rugelii*.

Lawrenceville, Lawrence County, June 27, 1926. Acc. No. *19477* (type).

This distinctive species forms the fourth of a group now known *Plantago*, among which it lies at the lower extreme of variation with respect to the size of the pycnidium. Upon this character alone, it can be separated very certainly from the others. In length of spore it shows more variation that *P. plantaginicola* Tehon & Dan., and lies midway between *P. plantaginella* Sacc. and *P. Plantaginis* Sacc. In spore width it coincides with the two last named, and approaches in its wider variates the narrower variates of the one first named.

Phyllosticta podophyllina Tehon & Stout, n. sp.

Foliicolous. Spots diaphyllous, circular, ½–4 mm. in diameter, tan, with a narrow, distinctly raised, concolorous margin;

the base of the spot collapsed and disorganized but not deciduous. Pycnidia abundant, scattered, spherical, developed in and occupying the mesophyll, visible amphiphyllously but opening epiphyllously, only the upper half of the pycnidium becoming erumpent, 70–95 μ in diameter. Ostiole circular, not papillate, 10–12 μ in diameter. Spores continuous, hyaline, oblong, with abruptly rounded ends, 6–8.5 × 2–2.5 μ.

On *Podophyllum peltatum*.

Columbia, Monroe County, June 24, 1926. Acc. No. *19480* (type).

The distinctive character of this species, as compared with *P. Podophylli* (M. A. Curt.) Wint., which we have collected and examined many times in Illinois, is as apparent when it is seen in the field as when it is dissected beneath the microscope. Microscopically, its pycnidia are smaller, and the spores are very characteristic, there being no possibility of confusing their distinctly oblong outline with the subcircular outline of the spores of the older species.

Phyllosticta allegheniensis Tehon & Stout, n. sp.

Foliicolous. Maculae diaphyllous, circular, 1–4 mm. wide, with a dark, purple-tinted, unraised margin about ½ mm. wide, tan to cinereous, similarly colored above and below. Pycnidia developed in the mesophyll, immersed, few, scattered, opening either upward or downward by a papillate ostiole, immersed portion brown and membranous, erumpent portion dark brown or carbonous, round or oval in outline, flask-shaped, and often applanate at the base, 90–130 μ in diameter; ostiole round to oval, 14–30 μ in diameter. Spores hyaline, oval, continuous, 2–2.5 × 4–4.5 μ.

On *Rubus allegheniensis*.

Nashville, Washington County, July 29, 1926. Acc. No. *20940* (type).

This seems to be different from other species noted on *Rubus*. From the two known American forms, it is distinguished as follows:

Spores minute, 5 μ or less long.
  Spores rod-like, 1–1.5 × 4.5–5 μ ..... *P. Dearnessii*
  Spores oval, 2–2.5 × 4–4.5 μ ..... *P. allegheniensis*
Spores larger, 5–7 μ long ..... *P. variabilis*

Phyllosticta subeffusa (Ellis & Ev.) Tehon & Stout, comb. nov., descr. emend.

Synonym: *Phyllosticta Smilacis* Ellis & Martin var. *subeffusa* Ellis & Ev. in Ellis & Everhart's North American Phyllostictas, p. 72, 1900.

Foliicolous. Spots very extensive, involving large areas of, or very often the entire, leaf, most often appearing marginal but in reality arising from a laminar infection, arid and of a dead gray closely simulating natural death; borders indefinite, usually brown- to red-tinted and shading into the natural green of the leaf. Pycnidia abundant, scattered, 85–105 μ in diameter, membranous, becoming subcarbonous at length, developed in and occupying the mesophyll, spherical to applanate, never erumpent but opening toward either side by a slightly papillate, irregular ostiole sometimes 40 μ wide. Spores hyaline to very dilute green, continuous, rather constantly elliptical in outline, 7.5–11 $\times$ 2.2–4 μ.

On *Smilax* sp. (West Va., Nuttall) Ellis & Ev. North American Fungi, No. *3252*.

Also on *Smilax hispida*.

Knoxville, Knox County, August 26, 1926. Acc. No. *19444*.

The writers have no hesitancy in making the above change in the nomenclature of this rare and interesting fungus. During the past 6 years we have seen, collected, and examined critically many specimens of *P. Smilacis*, without encountering anything that could be classed as the variety *subeffusa*. The striking character of the variety, as exhibited in the North American Fungi specimens, leaves little doubt of the type of infection to be found; but it required five years of searching for us to secure it in Illinois. A single vine, climbing over a small tree, bore a number of leaves with the characteristic spots, and subsequent examination proved them to be typical of the variety. Distinction may be found between the two forms not merely in the form and texture of the host lesions but also quite definitely in the fungi themselves:

Species	Pycnidial diameter	Spores		
		Length	Width	Shape
P. Smilacis	110–150 μ	12–15 μ	3.5–4 μ	Oblong-fusoid
P. effusa	85–105 μ	7–11 μ	2.2–4 μ	Elliptical

Phoma asparagina Tehon & Stout, n. sp.

Caulicolous. Maculicolous rather than cankerous, the spots at first showing as long-elliptic, slightly sunken, gray lesions, later becoming confluent and producing extensive gray patches from which the cuticle falls, often also with a reddish-brown to purple margin. Pycnidia abundant, scattered, located in the epidermis and partly exposed by the falling of the cuticle, globose or applanate, brown, membranous, 50–150 μ in diameter; ostiole papillate, dark and often carbonized, 10–30 μ in diameter. Spores continuous, hyaline, oblong-elliptic, 3.5–6 $\times$ 1–2 μ.

On *Asparagus officinalis*.

Anna, Union County, November 11, 1926. Acc. No. *19943* (type).

This and an ascomycete, listed on a previous page as *Metasphaeria Asparagi*, occur on the same spots and may be related conidial and ascigerous forms.

From the two previously described species of *Phoma* attacking asparagus, one European and the other American, our Illinois species is separated as follows:

Spores blunt; pycnidia small (150 μ in diameter).
Spores 7–8 $\times$ 3 μ ..... *P. Asparagi*
Spores 3–6 $\times$ 1–2 μ ..... *P. asparagina*
Spores acute; pycnidia ½ mm. in diameter ..... *P. media*

Macrophoma Smilacinae Tehon & Stout, n. sp.

Foliicolous. Spots circular to ellipsoid, about equally apparent on both sides of the leaf, 4–6 mm. long, sometimes confluent to form larger irregular lesions; their margins narrow, very definite, reddish-brown; their centers grayish-white and papery. Pycnidia scarce or many per spot and sometimes forming a circle near the periphery, macroscopically black, microscopically dark brown, membranous, of a meandering, plectenchymous structure, epiphyllous, flattened-globose, 100–225 μ in diameter; ostiole rounded, hardly papillate, but at least surrounded by a zone of darkened, thickened tissue, 12–22 μ across. Spores 1-celled, greenish to hyaline, irregularly narrow-ellipsoid, 11–22 $\times$ 3.5–6 μ, produced at the tips of hyaline sporophores.

On *Smilacina stellata*.

Marlow, Jefferson County, September 7, 1926. Acc. No. *20001* (type).

Macrophoma Cercis Tehon & Stout, n. sp.

Foliicolous. Spots measuring up to 8 mm. long, circular when small, becoming oblongate and somewhat angular by being limited by the leaf veins; the margin definite, dark brown; the inner part of the spot light tan. Pycnidia abundant, not gregarious, developed in and occupying the mesophyll, opening epiphyllously by a slightly papillate ostiole, flattened-globose, dark brown, composed of a densely interwoven hyphal structure (a sort of meandering plectenchyma), 110–185 μ in diameter; ostiole round or angular, sometimes showing a darkened rim when rounded, 12–15.5 μ across. Spores 1-celled, hyaline, ovoid to long-ellipsoid and sometimes somewhat irregular, 13–23 $\times$ 4.5–7.7 μ, produced at the tips of simple, definite, hyaline sporophores.

On leaves of *Cercis canadensis*.

Venedy, Washington County, September 8, 1926. Acc. No. *19972* (type).

Macrophoma Phlei Tehon & Stout, n. sp.

Foliicolous. No evident spots. Pycnidia appearing abundantly on dry, dead leaves from tip to sheath subsequent to fruiting of host, not gregarious but disposed in rows closely adjacent to the leaf veins, membranous, subcarbonous, or completely carbonous, developed in and occupying the mesophyll, rotund or applanate in longi-sections, often oval in outline, the long axis parallel with the vein when viewed from above, 105–225 μ in diameter. Erumpent hypophyllously only. Ostiole papillate, at first closed by a thin, cellular membrane which dissolves when the spores mature and leaves a very distinct, irregularly circular stoma 17–28 μ in diameter. Spores hyaline, continuous, oval, 18–26 $\times$ 6.4–7.7 μ.

On *Phleum pratense*.

Wayne City, Wayne County, November 8, 1926. Acc. No. *19413* (type).

This is, in all respects except spore color, similar to *Sphaeropsis Phlei* Ellis & Ev.; but in our specimen all the marks of maturity are present, including the ability of the spores to germinate. It seems proper, therefore, to record this as a distinct form.

Exophoma astericola Tehon & Stout, n. sp.

Colonies foliicolous, chiefly hypophyllous, gray, irregular or subcircular, 5–20 mm. in diameter or more extensive by con-

fluence. Mycelium abundant, chiefly external, hyaline, irregularly and copiously branched, 4–4.5 μ in diameter. Pycnidia abundant, entirely external, brown, membranous, subspherical to ovoid, astomatous until maturity, 35–77 $\times$ 22–45 μ, raised beyond the hyphal weft by a cellular stalk of variable length and width. Ostiole very variable, appearing as a rupture of the pycnidium's apex, often apparently but not truly rostrate, 10–14 μ wide. Spores 1-celled, hyaline to smoky, ellipsoid, 4–5 $\times$ 7–10 μ, usually covered with a perceptible gelatinous film.

On *Aster tardiflorus*.

Paris, Edgar County, November 4, 1926. Acc. No. *19386* (type).

Cyphellopycnis Tehon & Stout, n. gen.

Pycnidia much longer than broad, membranous to carbonous, of somewhat irregular outline, containing a single cavity which opens to the outside by numerous, irregularly placed, often confluent ostioles. Spores hyaline, 1-celled.

This genus, erected to contain the following species, belongs in the Phomataceae, but, as far as we can ascertain, represents a type of pycnidium not heretofore noticed. The pycnidial structure houses a unified cavity, in which there is no sign of the locular separation which would naturally be expected from the numerous ostioles.

Cyphellopycnis Pastinacae Tehon & Stout, n. sp.

Caulicolous. Not maculicole. Pycnidia immersed, discrete, in linear series between the sclerenchyma fibers of the stem, spherical, oval, or elongate, 400 up to 2,000 μ long, outline irregular. Ostioles several to many, erumpent, 50 to 75 μ or more wide, separate or confluent, and definitely delineated by a dark border. Spores hyaline, oval to ellipsoid, usually very distinctly biguttulate, 7.7 $\times$ 2.2 to 13.2 $\times$ 2.4 μ but ordinarily 8.5–11 $\times$ 2.2 μ. PLATE 13, FIG. 5.

On *Pastinaca sativa*.

Arnold, Morgan County, July 20, 1926. Acc. No. *13257* (type).

Cytospora sambucicola Tehon & Stout, n. nom.

Synonym: *Cytospora sambucina* Tehon & Daniels, Mycologia **19**: 122. 1927.

Not *Cytospora sambucina* Ellis & Barth. Erythrea **5**: 48. 1897.

The kindness of Elam Bartholomew prompted him to bring to our notice our unfortunate and erroneous duplication of name in "Notes—III." From our description, Mr. Bartholomew was of the opinion that we had renamed the species named by him and Mr. Ellis 30 years before and he presented us a portion of his original material with which to make comparison. We have studied both; and it seems to us that our more northern form ought to be regarded as distinct. In this, as in most cases of plant-inhabiting *Imperfecti*, it is exceedingly difficult to express in words the differences that are so evident to our eyes and that our experience tells us to value far more than the differences in measurements upon which we seem so confidently to depend. We may, however, append the following tabular comparison.

	Ostiolar Opening	Spores
C. sambucicola	Compound	4–6.5 μ long, 1–2.5 μ broad
C. sambucina	Simple	6–7 μ long, 1.25 μ broad

Diplodia acericola Tehon & Stout, n. sp.

Foliicolous. Spots diaphyllous, yellow to tan at first, dark brown at length, circular, 4–12 mm. in diameter; margin chocolate brown, ½ mm. wide, crenulate because limited by the veinlets. Pycnidia numerous, scattered, situated in the mesophyll, membranous but becoming dark and carbonous in the upper half, 150–195 μ in diameter, opening epiphyllously by a somewhat raised but hardly rostrate ostiole 12–16 μ wide. Spores dark green, oblong, 1-septate, the septum very dark and distinct, 19.8–26.4 × 8.8–13.2 μ.

On *Acer saccharum*.

Mt. Pleasant, Union County, July 7, 1926. Acc. No. *14104* (type).

The eight species of *Diplodia* hitherto noted on *Acer* fall into two general groups separable on the basis of their spore lengths, and the species themselves can be distinguished as follows:

Spores less than 20 μ long.
  Spores 10 to 15 μ long.
    Pycnidia disposed in linear series; spores 6–8 μ wide..*D. subtectoides*
    Pycnidia scattered; spores 4–5 μ wide.....*D. microsporella*
  Spores 17 μ long, 9 μ wide.....*D. acerina*
Spores 20 μ or more long.
  Spores not over 25 μ long.
    Pycnidia disposed in linear series.....*D. subtecta*

Pycnidia scattered.
Pycnidia "very minute" ..... *D. minutissima*
Pycnidia larger.
Septum of spore very dark, distinct, and pronounced. *D. acericola*
Septum and spore wall concolorous.
On box elder ..... *D. atrata*
On other maples ..... *D. petiolarum*
Spores more than 25 μ long ..... *D. extensa*

Cryptostictis inaequalis Tehon & Stout, n. sp.

Foliicolous. Pycnidia numerous, scattered, erumpent, spherical, dark but membranous, 90–150 μ in diameter; ostiole not rostrate, circular, usually enclosed by a carbonous ring, 14–25 μ wide. Spores 3-septate, the central septum usually not in the middle of the spore, end cells shorter than the middle cells, the spore 11–16 $\times$ 2–2.5 μ. Cilia 1 at each end cell, not apical, 11–15 $\times$ 0.25–0.5 μ, hyaline.

On *Vitis rotundifolia*.

Murphysboro, Jackson County, August 23, 1926. Acc. No. *13698* (type).

Though it appears from our specimen that this fungus gains entrance to the plant it inhabits by making use first of dead tissue killed by the black rot organism (*Guignardia Bidwellii*), it is to be remarked that the black rot organism has not been able to fruit in spots occupied by the *Cryptostictis*. Spores of *C. hysterioides* Fuckel., reported on *Vitis* in Europe, are said to be 16 $\times$ 7 μ and without cilia.

Septoria Tecomaxochitl Tehon & Stout, n. sp.

Foliicolous. Spots diaphyllous, small, 0.25–1 mm. in diameter, subcircular, tan to cinereous, with a broad, purplish, diffused halo above but not below. Pycnidia few, 1 to 4 or rarely 5 per spot, scattered, located in the palisade and epidermis, spherical, 50–90 μ in diameter, brown, membranous, protruding by means of a slightly papillate, somewhat carbonized ostiole with a round aperture at first 10–18 μ in diameter but later widely opened. Spores hyaline, straight to slightly curved, filiform, with no visible septa, 1–1.5 μ wide by 30–44 μ long.

On *Tecoma radicans*.

Lawrenceville, Lawrence County, October 26, 1926. Acc. No. *20946* (type). Hardin, Calhoun County, September 18, 1926. Acc. No. *20958*.

Our species differs quite obviously from *S. Tecomae* Ellis & Ev., not only in its distinctly larger pycnidia, which are as a rule distinctly larger than those of the Ellis species, but also with respect to its spores, which are both shorter and narrower.

Pseudodictya Tehon & Stout, n. gen.

Leptostromataceae. Pycnidia dimidiate, separate, membranous or carbonous, more or less superficial. Spores dark, septate, spherical to somewhat elongate. Epispore smooth.

Based upon the following remarkable species, this genus appears to fall in the Leptostromataceae, and is most readily disposed of by aligning it with the Phaeophragmiae, though its spore characters are not those usually expected in that group.

Pseudodictya sassafrasicola Tehon & Stout, n. sp.

Foliicolous. Spots circular, diaphyllous, reaching diameters of 3.5–6.5 mm., light brown throughout with a somewhat diffused, dark brown to black margin. Pycnidia abundant, widely scattered, strictly epiphyllous, developed and remaining between the cuticle and the epidermis, round, dimidiate, membranous when young, completely carbonized when old, astomous, 135–180 μ in diameter, 20–28 μ high. Spores brown, spherical, globose, or elongate-globose and falcate, 2-septate, 8.5–11 μ wide, 8.5–11 μ in length. PLATE 13, FIG. 9.

On *Sassafras variifolium.*

Seymour, Champaign County, October 15, 1925. Acc. No. *9353* (type).

The peculiar shape and septation of the spore of this fungus is misleading when first seen under the microscope. Usually it rests on the longer surface and appears as a globose body with muriform septations, but when it is rotated under the cover glass it is seen to have three distinct cells, placed end to end. The center cell is much the largest and, of the remaining two, one is distinctly larger than the other. This lateral view reminds one of the spores of *Helminthosporium inaequalis.*

Leptothyriella Liquidambaris Tehon & Stout, n. sp.

Foliicolous. Spots diaphyllous, brown, circular, 1–5 mm. in diameter, concolorous, collapsed, fragile, crumbling with age. Pycnidia sparse, epiphyllous only, superficial, dimidiate, radiate, 91–112 μ in diameter, without an ostiole, but with a circular

central "cell," 10–14 μ in diameter, from which the hyphae of the pycnothyrium seem to spring. Spores oval to oblong, virescent-hyaline, non-septate, 8.4–10.2 × 6–6.8 μ. PLATE 13, FIG. 7.

On *Liquidambar styraciflua*.

Olmstead, Pulaski County, August 9, 1922. Acc. No. *1445* (type).

We can not be wholly satisfied with the present disposition of this fungus. The "central cell," referred to in the description as being the point from which the pycnothyrial strands emanate, is apparently the apex of a stalk which connects with the internal mycelium and from which the sporophores are derived also. The entire group of pycnothyrial forms needs careful study.

Diplopeltis sassafrasicola Tehon & Stout, n. sp.

Foliicolous. Spots diaphyllous, very irregularly circular, 3–10 mm. in diameter, dark brown above when young, with a distinct, conspicuous, purplish marginal line, turning to a faded tan or cinereous, becoming friable and falling away with age, less distinct hypophyllously where the bloom of the leaf obscures the character. Pycnidia subcuticular, epiphyllous, few per spot, sparsely scattered, black, distinctly carbonized and impervious to transmitted light, circular in outline, in longitudinal section very flatly dimidiate, 120–270 μ in diameter, up to 64 μ high. Ostiole 7.5–14 μ in diameter, round, indistinct until the spores mature. Spores brown, 1-septate, typically oblong, with rounded ends, not perceptibly constricted, 18.5–22 × 7.5–11 μ. PLATE 13, FIG. 8.

On *Sassafras variifolium*.

Thebes, Alexander County, July 17, 1922. Acc. No. *581* (type).

The fungus is referred to *Diplopeltis* with hesitation. There are no characters to exclude it, if the Engler & Prantl characterization is followed; but if the characterization of the genus, and of the one previously described species, as given by Saccardo in the Sylloge is followed, it is doubtful whether this can be admitted, for the pycnidial cover, though parenchymatic, bears no indication, even in young pycnidia, of a radiate development.

The following species are being recorded either for the first time for Illinois or for the first time since the early collections

made in the State from about 1880 to 1890 by A. B. Seymour, F. S. Earle, M. B. Waite, and G. P. Clinton, and are represented by specimens deposited in the Mycological Collection of the Illinois State Natural History Survey.

Erysiphe Martii Lév. has been taken on *Urtica gracilis* near Fairview, Mason City, Rock Falls, and Wayne. These localities seem to indicate a state-wide distribution, though the fungus evidently is not common.

Mycosphaerella lethalis Stone, on *Melilotus alba*, is represented by a single specimen taken near Pearl City in the extreme north of the State.

Melanopsichium austro-americanum (Speg.) G. Beck. has been taken on *Polygonum pennsylvanicum* near Springfield.

Ustilago Cenchri Lagerh. has been collected once on *Cenchrus carolinianus* in the Illinois River sands near Beardstown.

Ustilago hypodytes (Schlecht.) Fries, collected by Clinton on *Stipa spartea*, has been found near Minier on *Stipa avenacea*.

Ustilago Rabenhorstiana Kuhn has been found on *Digitaria sanguinalis* near West Union. This fungus was collected in 1882 by Seymour on *Panicum glabrum* (*D. humifusa*) in 2 localities in northern Illinois, and on *Panicum sanguinale* in 1881 at Anna, Cobden, Monticello, Twin Grove, and in Henry County.

Ustilago Panici-glauci Wint. has been found in the vicinity of Urbana on *Setaria glauca*.

Urocystis Agropyri (Preuss.) Schroet. has been collected on *Elymus virginicus* near Atlanta, DeSoto, and Onarga. The increasing abundance of this grass along roadsides will probably be followed by an increased abundance of the smut.

Coleosporium Viburni Arth. (II) has been taken on *Campanula americana* near the towns of Henry and Mapleton.

Puccinia angustata Peck, hitherto found on *Scirpus cyperinus* (M. B. Waite, 1889) and *S. atrovirens* (A. B. Seymour, 1881, and M. B. Waite, 1885), has been found also on *S. polyphyllus* near Stronghurst.

Puccinia canaliculata (Schw.) Lagerh., reported by Kern (Mycologia **11**: 134. 1919) as occurring on *Cyperus strigosus*, has been collected in the vicinity of Thompson on *C. Schweinitzii*.

Puccinia Bardanae Corda has been found on *Arctium lappa* in

seven localities ranging from southern to extreme northern Illinois.

Phyllosticta sorghina Sacc. has been taken on *Holcus Sorghum* (broom corn) near Mattoon.

Phyllosticta Sassafras Cooke has been found near Marshall on *Sassafras variifolium*.

Phyllosticta orobella Sacc. has been found twice along the Illinois River, once on an unnamed *Lathyrus* and once on a legume not determined further by the collector.

Septoria sonchifolia Rob. & Desm. has been taken on *Sonchus oleraceus* at Colona, DuQuoin, and Fisher. The wide separation of these stations indicates a general distribution of the species.

Septoria Agrimoniae Roum. has been collected on *Agrimonia* sp. at Lilly, Aledo, and Mt. Sterling. The three stations are all in the northwest quarter of the State.

Septoria Agropyri Ellis & Ev. has been found on *Agropyron repens* at Lebanon and Mt. Carroll.

Septoria bacilligera Wint. has been identified on *Ambrosia trifida* at East Peoria, Kampsville, and McLeansboro—another species of wide distribution but of comparatively rare occurrence.

Septoria Bromi Sacc. has been collected on *Bromus secalinis* at Chester, Equality, Kampsville, Ridgway, Sparta, Waterloo, and White Heath and on *Elymus virginicus* at Arthur and Bement.

Septoria Brunellae Ellis & Holw. has been taken on *Prunella vulgaris* at 21 stations, reaching from the southern to the northern boundary of the State.

Septoria Campanulae (Lév.) Sacc. has been taken on *Campanula americana* at four stations and seems to have a range occupying a region just south of the middle of the State.

Septoria Commonsii Ellis & Ev. has been taken on *Cirsium lanceolatum* at three stations, all in the northern half of the State.

Septoria conspicua Ellis & Mart. has been taken on *Steironema ciliatum* at five widely separated stations and once on *S. lanceolatum* at Cisne.

Septoria Pileae Thuem. has been found on *Pilea pumila* at Lawrenceville.

Septoria Physostegiae Ellis & Ev. was taken on an undetermined species of *Physostegia* at Oregon.

Gloeosporium septorioides Sacc. has been collected on *Quercus alba*, *Q. imbricaria*, and *Q. macrocarpa*. The five stations represented by our specimens seem to indicate that this fungus is far more common in the southern than in the northern half of the State.

Gloeosporium fraxineum Peck has been taken once on *Fraxinus quadrangulata* at Oregon.

Gloeosporium Equiseti Ellis & Ev. has been collected on *Equisetum arvense* at Morton, Oregon, and Rochelle, and is apparently entirely northern.

Gloeosporium musarum Cooke & Massee, on *Musa sapientum*, has been taken at Mt. Carmel and Mt. Vernon.

Cladosporium Triostei Peck, on *Triosteum aurantacum*, has been collected at Stronghurst.

Cladosporium Pisi Cooke & Massee, on *Pisum sativum*, has been found at Normal.

Septogloeum subnudum Davis, on *Smilax hispida*, has been collected in the woods along the banks of the Sangamon River near Seymour.

ILLINOIS STATE NATURAL HISTORY SURVEY,
URBANA, ILLINOIS

EXPLANATION OF PLATE 13

Fig. 1. *Stigmatophragmia sassafrasicola*. An ascus, with spores and paraphyses.

Fig. 2. *Melanospora interna*. A spore, showing hyaline ends and the coarse, irregular reticulation of the wall.

Fig. 3. *Metasphaeria Asparagi*. An ascus, showing the shape, the thick apical wall, and the loose arrangement of the 4-celled spores.

Fig. 4. *Pleospora Oleraceae*. Two spores, showing septation and variation in size.

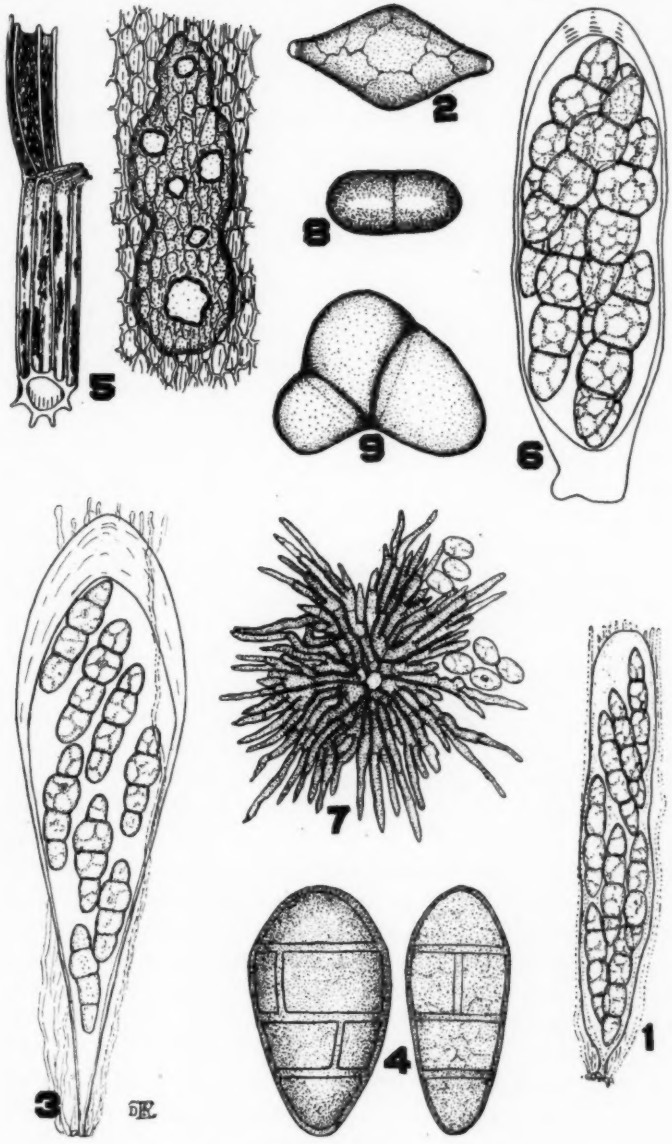
Fig. 5. *Cyphellopycnis pastinacae*. Habitat sketch, showing the fungus in blotches on a piece of a parsnip stem, and a typical pycnidium with several ostioles of varied sizes.

Fig. 6. *Metasphaeria sassafrasicola*. An ascus, showing the shape, the large foot, the double wall, and the arrangement of spores.

Fig. 7. *Leptothyriella Liquidambaris*. Pycnothyrium, showing the radiate structure and the relative size of the spores which issue from beneath its edge.

Fig. 8. *Diplopeltis sassafrasicola*. A spore, showing shape, septation, and very slight constriction.

Fig. 9. *Pseudodictya sassafrasicola*. Side view of spore, showing the large middle cell and the unequal end cells.



PARASITIC FUNGI OF ILLINOIS

NEW MEDIA FOR DEVELOPING SPOROPHORES OF WOOD-ROT FUNGI

BESSIE E. ETTER

(WITH PLATES 14 AND 15)

INTRODUCTION

For several years the writer has been experimenting with various culture media which would produce typical sporophores of wood-rotting fungi under control conditions in the laboratory. Long and Harsch [1] were able to grow many wood-rotting fungi in test tubes or small flasks containing agar decoctions; although fruiting bodies with typical hymenia and spores were often obtained, these were invariably undersized and in most instances without true pilei. Many of these experiments were repeated and a large number of sporophores obtained similar to those reported by Long and Harsch. In addition to this, sporophores with true pilei were grown inside small flasks. In every case the production of such sporophores was limited to fungi having a central stipe and a pileus which is normally not more than a half to one inch in diameter, such as *Polyporus perennis*, *Coprinus micaceus* and other similar fungi. However, the pilei thus produced, while of full size, did not have the typical zones and markings that characterize these species when grown in the open.

Small sporophores were also produced in flasks or test tubes of fungi which have large sporophores, such as *Pleurotus ostreatus*, *Lentinus lepideus*, etc., but such pilei were entirely lacking in the specific characters which determine these species when grown in nature. In the course of these experiments hundreds of small sporophores of wood-inhabiting fungi have been grown in flasks and test tubes, but rarely did such pilei have any markings or zones which would identify the species. The experiments so

[1] Long, W. H., and Harsch, R. M. Cultures of wood-rotting fungi on artificial media, Jour. Agr. Research **5**: 33–82. January 1918.

far conducted indicate that typical, well-marked, pileate sporophores of most fungi can not be developed inside containers. It seems that the zones, whether they be of hairs, scales or colors, must have open air conditions for normal development.

CULTURE MEDIA

It was soon found that the ordinary agar decoction, even in as large a vessel as a liter flask, apparently did not contain enough nutriment to produce typical sporophores of the larger fungi. Of course by using very large containers, which in an ordinary laboratory would not be practicable, sporophores of normal size might be produced, but even then the zones and markings would not be characteristic of the species in question. Therefore, experiments were begun with more or less solid media, such as sawdust, ground wood, corn-meal, starch, etc. Not only was a sufficient quantity of the solid nutrient necessary, but a more or less porous condition had to be maintained in the medium after sterilization; otherwise the hyphae would be limited mainly to the surface and sub-surface layers of the medium where they often could not obtain enough food to produce a large sporophore. It has been very difficult to obtain media sufficiently porous and yet containing enough moisture to keep the fungi in vigorous growth. In many instances the fungus would soon over-run the surface and produce a dense mat of mycelium which seemed to prevent the formation of sporophores.

To obtain the porous condition necessary many different materials were tried but so far none has been found that could be called a complete success for all species of fungi. Sponges soaked in various agar decoctions were tried but in every instance the heat of the autoclave, during sterilization, flattened the sponge and destroyed the porous condition which was so desired. Then blotting paper, filter paper, etc., were tried by making rolls in which there were alternate layers of the solid media and the porous paper soaked with liquid, but for some reason the fungous threads did not readily penetrate the layers of porous paper. Sphagnum moss mixed in varying proportions with the solid media was the best material found for increasing the porosity.

Various combinations of solid materials have been tried with more or less success; however, no medium was found which contained enough available initial liquid food to bring a large sporophore to maturity. It is, therefore, necessary to add liquid to the media after the cultures have been growing for a certain length of time.

For many wood-rotting fungi on conifers a very satisfactory culture medium for producing large sporophores is the following:

Corn-meal (white Quaker)	48 grams
Corn-starch (Kingsford)	16 grams
Powdered pine wood (*Pinus edulis*)	8 grams

To these solids is added a certain amount of malt liquid, made by adding 25 grams of malt extract to one liter of water. The quantity of this liquid to be added will of course be determined by the amount of solids used and the size of the container in which the fungi are growing. The malt preparation should not only saturate the solids used but should fill the flask about $\frac{2}{3}$ full, leaving the upper $\frac{1}{3}$ of the solids exposed to the air in the flask.

Flasks from 250 to 500 cc. were used. A 250 cc. flask was rather small; so the 500 cc. was generally used, although well developed, full size, characteristic sporophores of *Lentinus lepideus* were obtained by the use of either size flask. In some of the tests no sphagnum moss was added to the media (PLATE 14A) while in others this moss was used (PLATE 14B). In either case good sporophores were obtained. If the sphagnum is used the 500 cc. flasks are necessary in order to obtain the amount of nutrient material for the full development of the sporophores since the sphagnum occupied about one half of the space.

Cane-sugar was substituted in some cases for the corn-starch but with poor results. The main difficulty encountered in the use of the corn-starch was the swelling and gelatinizing of the starch granules, thus tending to destroy the desired porosity of the media. Any coniferous wood can be used if it is soft enough to produce a fine powder or flour. The powdered sap-wood of Western yellow pine (*Pinus ponderosa*), Mountain white pine (*Pinus flexilis*), Pinon (*P. edulis*), and Engelmann spruce (*Picea Engelmanni*) have been used with success. Powdered cotton-

wood was used instead of powdered pine for the production of the sporophores of fungi which attack hardwoods, the remainder of the formula being the same as that used for the conifers. For the growing of sporophores from fungi which attack certain forms of wood such as junipers, red-wood, oaks, etc., the powdered wood of these species in question was used.

PREPARATION OF THE POWDERED WOOD

The most important thing is to have the wood in as *finely powdered condition* as possible so that it will be readily and quickly available for fungous growth when a sufficient amount of moisture is present. The wood is powdered by holding it against a rapidly revolving disc driven by an electric motor. Coarse sandpaper is attached to the surface of the disc. Garnet sandpaper No. 2½ was found the most satisfactory since this paper was fine enough grained to produce a dust-like powder, but was coarse enough so that any oils or resins present in the wood would not gum the sandpaper and thus stop the powdering action. Several kinds of wood flour were made in advance and kept in paper boxes properly labeled.

In the earlier experiments one half of the media was wood powder but this large amount was found unnecessary for producing the desired sporophores and was very wasteful of the powdered wood. This last is an important item, for the powdering of the wood is a tedious and time-killing process. The amount of powdered wood given in the present formula was found sufficient to produce large sporophores of the various fungi tested. The solid portions of the media consisting of corn-meal, starch, and wood powder were thoroughly mixed in considerable quantities and kept in paper boxes ready for use at any time. The mixture, in the dry climate of Albuquerque, keeps perfectly without any signs of molding or mildewing. In damper climates it might be necessary to keep it in glass containers with airtight tops, like candy jars.

STERILIZATION OF MEDIA

After the proper amount of the malt liquid is added to the solid portions of the media, the flasks are plugged with cotton

in the usual manner and sterilized in an autoclave at about 8 lbs. pressure, for at least one hour. The flasks must not be too full; otherwise the swelling of the solid portions of the media during sterilization and the often uneven pressure between the inside and the outside of the flask will cause the media to wet the plugs or even push them out.

THE ADDING OF THE MALT LIQUID

Flasks proved satisfactory when the fungi used had well pronounced stipes. The method of procedure in such cases was as follows: The flasks with the proper amount of media in them are first plugged and sterilized and then inoculated with the fungi to be tested. As the fungus develops more liquid medium is added when necessary. It is important that this food be added at the proper time and with extreme care, to prevent contamination. The most satisfactory method of adding this malt was found in the use of large test tubes 25 by 200 mm. in size. These were filled about ⅔ full of the malt liquid, then plugged and sterilized in the usual manner. Whenever the liquid food was added to the cultures the mouths of the tubes and of the culture flasks were flamed and the entire contents of the test tube were poured into the culture.

The time at which the malt should be added to the cultures is of vital importance and is one that can be determined only by observation and experience. This varies with the fungus under investigation and often several trials must be made before the proper time can be determined. With typical stipitate sporophores, the best time to add the liquid media is usually when the young and growing stipe has become clearly differentiated and is about an inch tall. The disappearance of the liquid in the flask and the slowing down of the growth of the ascending stipe indicate when more malt is needed. Two or three additions of malt are usually sufficient to bring the sporophore to full maturity.

DEVELOPMENT OF THE SPOROPHORES

The growth of the sporophore in the flask usually proceeds very rapidly and the ascending stipe soon reaches the cotton plug. At this stage the plug is withdrawn, spread over the top

of the flask and held in place by rubber bands or strings. Of course, the sterilized portion of the plug after spreading should still be above the mouth of the flask. It is an easy matter for the ascending sporophore to push its way through the cotton covering into the outside air. The pileus will usually expand very rapidly if a sufficient amount of liquid medium is maintained in the flask. At this stage, the surface of the culture in the flask will be so thoroughly matted with felts of mycelia that no visible contamination will appear until long after the sporophore has completed its growth, discharged its spores and become dry.

The length of time between the inoculation of the culture media and the formation of perfect sporophores varies with the fungus. Perfect sporulating sporophores of *Lentinus lepideus* (PLATE 14) were obtained in from six to eight weeks after inoculation; with *Pleurotus ostreatus* this period was somewhat shorter. Typical pileate sporophores of the following wood rotting fungi have been grown on the media described: *Lentinus lepideus*, *Pleurotus ostreatus*, *Coprinus micaceus*, *Coprinus atramentarius*, *Xerotus* sp., *Pholiota* sp., *Polyporus arcularius*, *Polyporus perennis*, *Polyporus Farlowii*, *Ganoderma Curtisii*, *Ganoderma polychromum*, *Ganoderma* sp. and *Trametes Peckii*. The stipitate forms were grown by the flask method described in this paper, while the bracket fungi were grown on the same media but by a different method, which will be discussed in a later paper.

SUMMARY

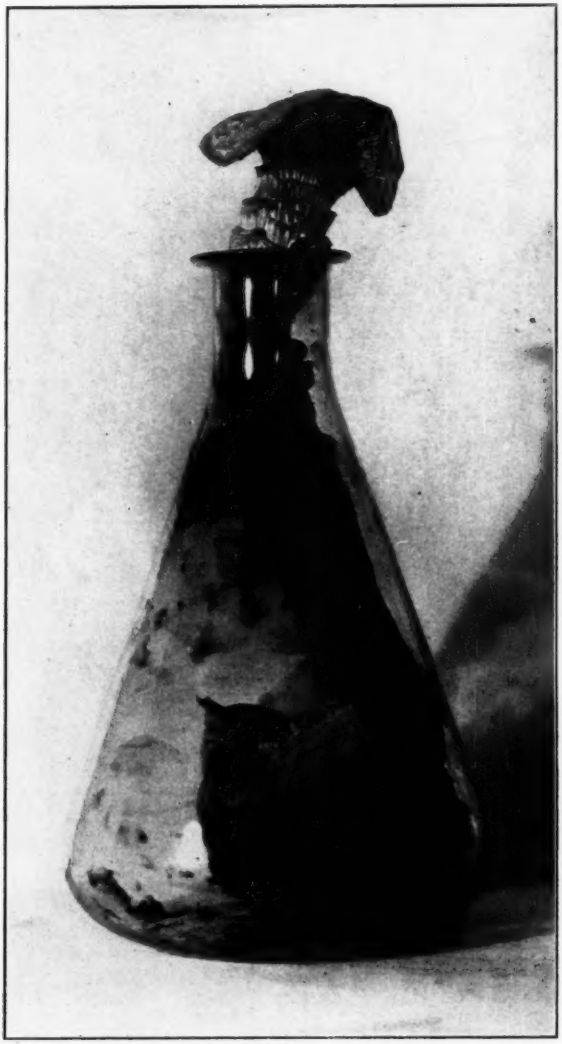
1. Media have been developed which produced characteristic pilei, normal as to size and markings, of the wood rotting fungi tested.

2. The media used consisted of mixtures of corn-meal, starch, wood flour and malt liquid.

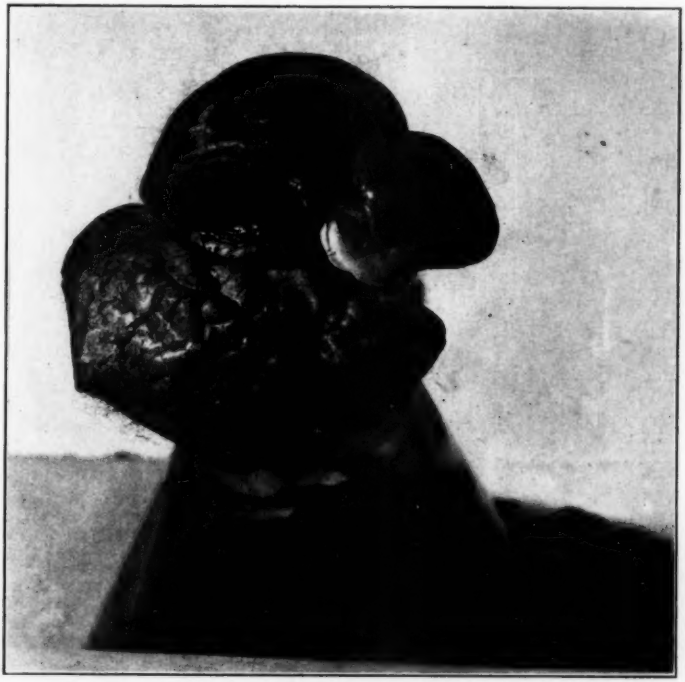
3. The cultures grew rapidly on these media and soon developed typical sporophores.

4. Media for growing sporophores of the larger fungi apparently must contain wood in some form in order to produce pilei of normal size.

5. Wood flour is the best of the materials tested since it is immediately available for use by the growing fungi.



LENTINUS LEPIDEUS (× 9/10) IN 250 CC. FLASK WITH NO SPHAGNUM MOSS IN MEDIUM. SHOWS TYPICAL MARKINGS ON STIPE



LENTINUS LEPIDEUS (× 9/10) IN 500 CC. FLASK WITH SPHAGNUM MOSS ADDED TO MEDIUM. SHOWS CHARACTERISTIC MARKINGS ON SURFACE OF PILEI.

6. Liquid media must be added to the growing cultures to produce normal pilei of fungi which have large sporophores.

7. Pilei normal as to size and markings would not develop *inside* containers of ordinary size.

8. The sporophores must extend beyond the mouth of the container into the outside air to produce characteristic pilei.

9. The flask method described is especially adapted to fungi having stipitate sporophores.

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CONTRIBUTIONS TO A MYCOLOGICAL FLORA OF LOCAL SOILS

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(WITH PLATES 16–19)

This investigation has been undertaken with the purpose of initiating observation of the fungous flora of the soils of Illinois. Studies of this nature have been made in a number of states, but none has been reported from Illinois up to the time of the present one. This work was carried on in Evanston, Illinois, during the year 1927–1928 under the helpful supervision of Dr. Alfred H. W. Povah.

MATERIALS

The materials employed were sterile test tubes, petri dishes, a scalpel and an alcohol lamp. Soil samples were taken during the months of October and November, 1927, while the earth was still unfrozen, from the surface and at depths varying from five to one hundred and twenty centimeters. Complete collection data are recorded in Table I.

METHOD

Before taking a soil sample, a few cubic millimeters of soil were scraped away with a flamed scalpel to eliminate possible surface contamination. Where samples were secured from the side of a ditch, several cubic centimeters of soil were removed, in order to avoid surface and air fungi. After being flamed, the test tube was forced to a depth of three or four centimeters into the soil, withdrawn, and stoppered. Blakeslee's (**21**) malt extract agar was the principal medium used in the study, as it favored luxuriant growth of a large proportion of the species examined. In addition to agar, moist bread in wide-mouth 250-cc. bottles, which had been autoclaved for two hours at fifteen pounds steam pressure, was used for the cultivation of species of Mucorales. Sterile green beans were also used for the growth of a number of

Fungi Imperfecti in order that the most favorable development might be obtained.

TABLE I

SOIL COLLECTION DATA

No.	Date	Type of Soil [1]	Depth
1	9/30/27	Black, filled with small roots	5 cm.
2	9/30/27	Black, free of roots	15 cm.
3	9/30/27	Light-colored, sandy	25 cm.
4	10/ 3/27	Sand and loam, stony	60 cm.
5	10/ 3/27	Black, filled with grass roots	5 cm.
6	10/ 3/27	Dark, mixed with sand and some clay	60 cm.
7	10/ 3/27	Light-colored, loose, sandy	90 cm.
8	10/ 3/27	Dark, some clay	120 cm.
9	10/12/27	Dry sand from shore of Lake Michigan, 39 cm. from water's edge	Surface
10	10/12/27	Wet sand from beach, just at water line	Surface
11	10/13/27	Cinders mixed with sand	5 cm.
12	10/13/27	Sand with slight admixture of cinders	15 cm.
13	10/13/27	Sand with loose stones	45 cm.
14	10/20/27	Garden soil; humus containing leaf mold	Surface
15	10/20/27	Dark moist humus, uncultivated	Surface
16	10/22/27	Uncultivated leaf mold	Surface
17	10/24/27	Dark leaf mold, uncultivated	Surface
18	10/28/27	Rich uncultivated humus	Surface
19	11/ 3/27	Dark sandy, from canna bed	Surface
20	11/10/27	Sandy, from cultivated conifer bed	Surface
21	11/10/27	Dark humus, from area wooded with oaks	Surface
22	11/10/27	Dark, from uncultivated open meadow	Surface

In all cases, strict precautions were taken to avoid contamination of the media and instruments. All glassware was sterilized in a hot air sterilizer at 150° C. for a period varying from forty-five minutes to one and one-half hours. Instruments such as needles, spear-points, wire loops and scalpels were flamed before use. The greatest care was taken in culture work to prevent contamination by spores present in the air of the laboratory. Such work was done under a glass case the interior of which had been washed with mercuric chloride solution (1–1,000).

After a tube of soil had been brought into the laboratory, sterile water was poured into it until the soil was thoroughly moistened and approximately a centimeter of clear liquid remained above the soil surface. The tube contents were then shaken vigorously at intervals of five to ten minutes until the soil was well "washed." Allowing a few minutes for the coarser

[1] Samples below the surface were obtained from the sides of a freshly dug trench, in uncultivated soil.

particles to settle, while the lighter ones such as fungal spores were still suspended, a flamed wire loop was dipped into the partially cleared liquid, and the droplet thus obtained was touched at several points on a poured agar plate. This process was repeated a number of times with each sample, so that two to three petri dish cultures were made from every collection of soil. The plates were then set aside and the fungi allowed to develop. The laboratory temperature averaged 20–22° C. during the day, but at night and over week-ends it fell frequently to 10–15° C. From the petri dish cultures, pure single spore isolations were made by the dilution method.

Clements' *Genera of Fungi* (**6**) was used most extensively in the determination of genera of the forms isolated in this study. In the identification of Mucors the works of Lendner (**13**), Van Tieghem (**29**) (**30**) (**31**), Hagem (**9**) (**10**) and Povah (**21**) were used. The key of Thom and Church (**28**) was employed in the identification of *Aspergilli*. In connection with the Fungi Imperfecti the writer found Rabenhorst (**4**) (**15**) and Engler and Prantl (**14**) of valuable assistance. Saccardo (**25**) was used repeatedly for general reference.

The Fusaria isolated were not referred to species, due to the unorganized state of the genus, but were merely placed in the "sections" indicated by Wollenweber, Sherbakoff et al. (**36**).

The writer is indebted to Dr. Charles Thom for the identification of a number of species of *Penicillium*, grateful acknowledgment of which is made at this time.

RESULTS

A list of the species of fungi isolated, arranged alphabetically, is given in Table II, with an indication of the depth in the soil at which each was found.

In the pure sand from the beach of Lake Michigan, three of the higher fungi were found: *Rhizopus nigricans*, an *Actinomyces* (described as *Actinomyces* IV), and a pink yeast.

Several forms apparently belonging to the *Actinomyces* group were observed in the course of this study, but they were not identified. A macroscopic description of each is given. Several yeasts, distinguishable by their color, were also noted, but no attempt was made to identify them.

TABLE II

VERTICAL DISTRIBUTION OF FUNGI ISOLATED FROM SOIL

Species	Surface	5 cm.	15 cm.	25 cm.	45 cm.	60 cm.	90 cm.	120 cm.
Actinomyces I	.	.	.	.	.	*		
Actinomyces II	.	.	.	.	.	*		
Actinomyces III	.	.	.	.	*			
Actinomyces IV	*							
Alternaria humicola Oud.	**	*	.	*				
Aspergillus fumigatus Fres.	.	**	*					
A. luchuensis Inui	***	**						
A. niger Van Tiegh	***	**	**					
Chaetomium subterraneum Swift & Povah.	.	.	.	.	.	.	.	*
Circinella simplex Van Tiegh.	***					*		
Cunninghamella elegans Lendner	**							
Fusarium arthrosporiella Sherb.	.	*						
F. elegans Wollen.	***							
F. liseola (Sacc.) Wollen. (?)	**							
F. roseum Wollen.	***							
F. sp.	*							
Hormodendrum cladosporioides Fres.	***	**	*	*				
Mucor abundans Povah	**							
M. circinelloides Van Tiegh.	***							
M. griseo-cyanus Hagem	***							
M. griseo-lilacinus Povah	***							
M. varians Povah	***							
Myrothecium convexum Berk.	*							
Penicillium roseum Link	**							
P. pinophilum Hedgcock	***	**						
P. stoloniferum Thom.	.	.	*					
P. III	.	**	.	*	.	.	*	
P. IV	.	*	.	.	.	*	.	*
P. V (monoverticillati series)	.	.	.	.	*			
P. VI (expansum series)	**							
P. Herquei Bainier	***							
P. oxalicum Currie & Thom	*							
P. IX	**							
Rhizopus nigricans Ehren.	***	**						
R. nigricans var. minor Jensen	.	.	*					
R. nodosus Namysl.	**							
Stachybotrys atra Corda	.	.	.	.	*			
Stysanus medius Sacc.	.	.	.	*				
Trichoderma Koningi Oud	***	***	*	**	.	*	.	*
Trichosporium nigricans Sacc. f. lignicola	.	.	.	*				
Trichurus terrophilus Swift & Povah	.	.	.	*				
Verticillium lateritium Berk.	.	.	.	.	.	.	*	
Zygorrhynchus Vuilleminii Namysl.	***	***	.	**	.	*	*	
Z. Moelleri Vuill.	.	*						
Approximate totals	80	26	7	10	3	5	3	3

* one isolation.
** two–three isolations.
*** four or more isolations.

In a review of the results of the present study, the writer found species of *Mucor*, *Aspergillus*, *Penicillium* and *Trichoderma* occurring most frequently, with *Zygorrhynchus*, *Rhizopus* and *Fusarium* also very common.

Two new species of fungi have been isolated and described; namely, **Chaetomium subterraneum,** found at a depth of 120 centimeters (Soil Sample No. 8), and **Trichurus terrophilus,** occurring at a depth of 25 centimeters (Soil Sample No. 3).

SPECIES ISOLATED

Identification of the fungi isolated constituted the principal part of this study. Following is a classification of the species found, with notations as to their occurrence. The numbers given in connection with the source of each species refer to Table I. In determining colors, Ridgway's *Color Standards and Color Nomenclature* (**24**) was followed.

I. PHYCOMYCETES.

A. MUCORALES.

1. SPORANGIOPHORAE.

a. MUCORACEAE.

Circinella simplex Van Tiegh. (PLATE 16, FIGS. 3, 4.)

This species was isolated from five different soils, cultivated and uncultivated, all taken at the surface (Nos. 14, 15, 17, 20, 21). Previously unreported from the soil.

Mucor abundans Povah.

This species was isolated twice by the writer from surface samples of dark sandy soil of cultivated canna bed (No. 19). Povah found this form in sandy tilled soil in Michigan in 1916.

Mucor circinelloides Van Tiegh.

Isolated repeatedly from surface soil taken from cultivated and uncultivated areas (Nos. 14–21). Previously reported from the soil by Jensen in 1912, Dale in 1914, Povah in 1916, Waksman in 1917, Pratt in 1918, and Takahashi in 1919.

Mucor griseo-cyanus Hagem.

Isolated several times from sandy, cultivated surface soil of flower bed, as well as from humus of wooded areas (Nos. 14, 16). Previously reported from the soil by Lendner in 1908 and Hagem in 1910.

Mucor griseo-lilacinus Povah. (PLATE 17, FIGS. 11, 12.)

This *Mucor* was found repeatedly in samples taken from the surface of cultivated and uncultivated sandy and loamy soils (Nos. 14–21). Previously unreported from the soil.

Mucor varians Povah.

This species was the most abundant in the collection of the writer. It was isolated many times from all surface samples, excepting those from the beach and from the open meadow. In collections from uncultivated soil (Nos. 15–18) the variation in form of columella was markedly less than in the other isolations. This species was found in tilled and untilled soil by Povah in 1916.

Rhizopus nigricans Ehren.

Found often in surface soils and less frequently at depths of 5 centimeters (No. 1). It also appeared on cultures from beach sand, with an *Actinomyces* and a pink yeast. On bread it formed a very dark turf reaching a height of 1–3 cm. after two weeks of growth. Isolated from the soil by Adametz in 1886, Hagem in 1907, Jensen in 1912, McLean and Wilson in 1914, Werkenthin in 1916, Waksman in 1917, Pratt in 1918, Rathbun in 1918, Takahashi in 1919, and Abbott in 1923.

Rhizopus nigricans Ehren. var. *minor* Jensen.

This variety was isolated from sandy filled-in soil containing some cinders, at a depth of 15 cm. (No. 12). Previously reported by Jensen in 1912.

Rhizopus nodosus Namysl.

This species was isolated twice from cultivated surface soil (No. 14). Previously reported from the soil by Hagem in 1907, Lendner in 1908, Jensen in 1912, and Waksman in 1917.

Zygorrhynchus Vuilleminii Namysl.

This fungus was found to be very abundant in all surface soils, and was also present in soils taken at depths of 5, 25, 60 and 90 centimeters (Nos. 1, 3, 5, 6, 7). It was the only representative of Mucorales in the soils taken from the open meadow (No. 22). Previously reported by Namyslowski in 1910, Jensen in 1912, Povah in 1914, Waksman in 1917, and Abbott in 1923.

Zygorrhynchus Moelleri Vuill.

Found by the writer but once. Its source was 5 cm. below the surface in loamy soil (No. 5). Previously found in the soil by Hagem in 1907, Jensen in 1913, and Paine in 1927.

2. CONIDIOPHORAE.

a. CHAETOCLADIACEAE.

Cunninghamella elegans Lendner.

This species was isolated twice from garden soil (No. 14). Previously reported from the soil by Lendner in 1908, Jensen in 1912, and Povah in 1914.

II. ASCOMYCETES.

A. SPHAERIALES.

1. SORDARIACEAE.

Chaetomium subterraneum Swift & Povah, sp. nov. (PLATE 19, FIGS. 6–11)

Forming on Blakeslee's agar circular *colonies*, at first grayish white, then slate blue-green gray and slightly iridescent, becoming deep olive gray and at maturity dark olive gray, almost black; reverse deep olive green; *mycelium* at first hyaline, later olivaceous, septate, 2–4 μ in diameter, aggregated in rope-like strands from which brown perithecia arise; *perithecia* 150–275 $\times$ 70–100 μ, spherical when young, becoming ovoid or flask-shaped, uniformly covered with mostly simple, straight, attenuate, six- to nine-septate, dark brown *setae*, 52–105 $\times$ 3 μ; setae with bulbous base 4–5 μ in diameter, often with elbow-turn just at swelling, sometimes very slightly undulating in upper half; shorter setae 20–30 μ long, often surrounding the ostiole; *asci* when young clavate, with short hyaline stalk, evanescent, sporogenous portion 21–30 $\times$ 8–14 μ, eight-spored, uniseriate; *spores* 7–10 $\times$ 5–7 μ, lemon-shaped, dark olive green, often containing one or more large oil globules, when young greenish in color and containing droplets of refractive substance.

While the hairs in **Chaetomium subterraneum** are typically unbranched, careful examination of the lower portion of the perithecia showed the occasional occurrence of hairs once branched near the base. Chivers (**5**) reports the occurrence of only one species of *Chaetomium* with unbranched straight terminal hairs, *Chaetomium trigonosporum* Marchal, but the shape of spores and the measurements of perithecia, hairs and asci do not agree with the above-described species.

This fungus was isolated from clay-mixed soil taken at a depth of 120 centimeters (No. 8).

III. **FUNGI IMPERFECTI.**

A. HYPHOMYCETES.

1. MUCEDINEAE.

a. MUCEDINACEAE.

Aspergillus fumigatus Fres. (PLATE 17, FIGS. 4–6.)

Isolated from humus at depths of 5 and 15 centimeters (Nos. 1 and 2). Previously reported from the soil by Waksman in 1917, Takahashi in 1919, and Paine in 1927.

Aspergillus luchuensis Inui. (PLATE 17, FIGS. 1–3.)

This species is a subdivision of the *niger* group of Thom and Church (**25**), and, though not reported under this name, has no doubt been found before in the soil and identified as *A. niger*. It was found repeatedly in surface soils by the writer, and several times at a depth of 5 centimeters (No. 5).

Aspergillus niger Van Tiegh.

Found repeatedly in surface soils and also at depths of 5 and 15 centimeters (Nos. 2 and 5). Previously reported by Dale in 1914, Waksman in 1917, Rathbun in 1918, Takahashi in 1919, Abbott in 1923, and Paine in 1927.

Penicillium roseum Link (?). (PLATE 16, FIGS. 7, 8.)

Isolated three times by the writer from cultivated surface soil. Also reported from the soil by Takahashi in 1919 and Paine in 1927.

Penicillium pinophilum Hedgcock.

Found repeatedly in surface soil and at a depth of 5 centimeters (No. 1).

Penicillium stoloniferum Thom.

Found at a depth of 15 centimeters (No. 2).

Penicillium sp. (III).

Colony low growing; spores forming powdery patches, Lincoln green; submerged mycelium white; numerous bright terra cotta perithecia formed over upper surface of colony and especially abundantly along the margins; reverse of culture on Blakeslee's agar honey yellow.

This species was found several times at a depth of 5 centimeters (No. 5), and once at 25 and again at 90 centimeters (Nos. 3, 7).

Penicillium sp. (IV).

Colony low growing, with leaf green powdery surface; submerged mycelium white. A few testaceous (Ridgway) perithecia are formed in clusters along margin of the culture. Reverse of colony clay color.

This *Penicillium* was taken from depths of 5, 60 and 120 centimeters (Nos. 5, 6, 8).

Penicillium sp. (*monoverticillati* series) (V).

Colony dusty with Lincoln green spore mass, concentrically zoned; submerged mycelium hyaline and inconspicuous; reverse of culture honey yellow.

A single isolation was made from the soil taken at a depth of 45 centimeters (No. 13).

Penicillium sp. (*expansum* series) (VI).

Old cultures Andover green, showing concentric zonation; spore production abundant; mycelium white, inconspicuous; reverse predominantly wood brown shading into Hay's russet.

Found several times in surface samples (Nos. 16, 17).

Penicillium Herquei Bainier.

Found repeatedly in surface soils (Nos. 14, 15, 18–22).

Penicillium oxalicum Currie & Thom.

This species was found in surface soil (No. 15).

Penicillium sp. (near to *aurantio-violaceum* Biourge) (IX).

Surface powdery with irregularly heaped Lincoln green spore mass; somewhat concentrically zoned; mycelium white; honey yellow resinous droplets concentrically arranged, covering surface of small terra cotta perithecia found principally at base of culture tube; reverse bright liver brown or Hay's russet.

This form was found several times in surface soils (No. 16).

Trichoderma Koningi Oud. (PLATE 18, FIGS. 4, 5.)

Isolated repeatedly from samples taken at the surface and from depths of 5, 15, 25, 60 and 120 centimeters (Nos. 2, 3, 5, 6, 8). Also found in the soil by Oudemans and Koning in 1902,

Jensen in 1912, Goddard in 1913, Dale in 1914, Waksman in 1917, Rathbun in 1918, Takahashi in 1919, and Abbott in 1923.

Verticillium lateritium Berk. (PLATE 16, FIGS. 5, 6.)

Found by the writer at a depth of 90 centimeters in sandy soil (No. 7). Previously unreported from the soil. In view of the fact that this fungus causes a serious disease of the Irish potato, its isolation from non-agricultural soil may have a significant bearing on the control of this disease.

2. DEMATIEAE.

a. DEMATIACEAE.

Alternaria humicola Oud. (PLATE 18, FIGS. 1, 2.)

This species was isolated by the writer a number of times from surface soils and at depths of 5 and 25 centimeters (Nos. 3, 5, 14). Reported from the soil by Koning in 1902, Dale in 1914, Waksman in 1917, and Abbott in 1923.

Hormodendrum cladosporioides Fres. (PLATE 18, FIG. 3.)

Found repeatedly in surface soils and at depths of 5, 15 and 25 centimeters (Nos. 1, 2, 5). Previously reported by Jensen in 1912, Goddard in 1913, and Paine in 1927.

Stachybotrys atra Corda. (PLATE 18, FIGS. 6, 7.)

Found by the writer in sandy soil at a depth of 45 centimeters (No. 13). Previously reported by Jensen in 1912.

Trichosporium nigricans Sacc., f. *lignicola*, as given by Saccardo.

Found by the writer at a depth of 25 centimeters in sandy soil (No. 3). Not previously reported from the soil, but Saccardo (**25**) indicates that it was found in France on decaying wood in association with *Hypocrea rigens*. This form, as developing in the writer's culture, offered a striking resemblance to the imperfect stages of a number of Ascomycetes related to *Hypocrea* (**32**), and might well be found upon investigation to be a stage in the life history of *Hypocrea rigens*.

3. STILBEAE.

a. STILBACEAE.

Stysanus medius Sacc.

This species was found by the writer at a depth of 25 centimeters in sandy soil (No. 3). Not previously reported as occurring in the soil.

Trichurus terrophilus Swift & Povah, sp. nov.
(PLATE 19, FIGS. 1–5)

Forming on Blakeslee's agar irregular *colonies*, at first pale olive gray with radial folds, becoming dark olive gray and finally olivaceous-black, always with pale margin, at maturity forming a dense and powdery growth up to 1.5 mm. in height, with small light-refracting droplets often on the surface; reverse greenish black; *mycelium* dark brown, septate, 2–3.5 μ in diameter, in early stage forming branched catenulate conidia on single hyphae. At maturity the mycelium adheres in rope-like strands from which arise vertically dark clavate *fruit bodies* 375–1300 μ tall, on stalks 95–800 $\times$ 20–70 μ, the fertile portion of the fruit body 135–500 $\times$ 35–150 μ, giving rise to simple or panicled chains of spores interspersed with bristle-like dark brown *setae* 15–70 μ in length, 3 μ wide at base, tapering gradually almost to the apex, which is terminated in a sharp point. Setae non-septate, or occasionally with one or two septa near the base, simple or forked, the two branches commonly of unequal length and forming an obtuse angle. *Spores* oval to elliptical, 3–6 $\times$ 2–3.5 μ, pale green, greenish black in mass.

This species was isolated by the writer from sandy soil at a depth of 25 centimeters (No. 3). Only three species of *Trichurus* are known: *T. cylindricus* Clements and Shear (**25**), *T. spiralis* Hasselbring (**11**), and *T. gorgonifer* Bainier (**25**). **T. terrophilus** differs from *T. cylindricus* in its much smaller spores, and from *T. spiralis* and *T. gorgonifer* in the form and branching of its setae, as well as in the smaller size of the spores.

4. TUBERCULARIEAE.

a. TUBERCULARIACEAE.

Fusarium arthrosporiella Sherb. (Sec. 5).

This *Fusarium* was isolated from loam at a depth of 5 centimeters (No. 5).

Fusarium elegans Wollen. (Sec. 13).

This fungus was found by the writer in surface soils (Nos. 14, 15, 21, 22).

Fusarium liseola (Sacc.) Wollen. (?) (Sec. 8).

The fungus isolated differs from typical *F. liseola* in that chlamydospores are present. It was found in surface soil (Nos. 14, 15).

Fusarium roseum Wollen. (Sec. 7).

This species was found repeatedly in surface soils (Nos. 15–22).

Fusarium sp.

This form does not fall into any of the sections described by Wollenweber et al. (**36**). It is comparatively slow-growing, with little aerial mycelium except at the edges of colony; surface of culture at first dull white, later sulphine yellow and somewhat powdery; sparse aerial mycelium white, with traces of red appearing when in contact with test tube; *microconidia* irregularly rod-shaped, 6–14 × 3–6 μ, 0–2 septate, sessile, guttulate; *macroconidia* sickle-shaped, 25–58 × 5–8 μ, three- to seven-septate, attenuate at tips; *mycelium* 3–7 μ in diameter, septate, aggregating in rope-like strands.

This species was found by the writer in surface soil (No. 20).

Myrothecium convexum Berk. & Curt. (PLATE 17, FIGS. 7–10.)

This species was isolated by the writer from sandy surface soil of cultivated conifer bed (No. 20). Previously unreported from the soil.

IV. ACTINOMYCES.

Actinomyces I.

Colony irregular in outline, prostrate, dull white at first, becoming creamy with age, wrinkled and folded similar to bacterial growth; surface somewhat powdery. This form was found at a depth of 60 centimeters (No. 4).

Actinomyces II.

Colony circular, at first white, then bright coral pink with hyaline margin, wrinkled radially, with a few strands of aerial white mycelium, forming a tough leathery growth on the surface of the substratum.

This species was found at a depth of 60 centimeters (No. 4).

Actinomyces III.

Colony at first circular, becoming irregular, presenting general appearance of bacterial growth, cream white; surface with numerous fine sinuous folds at center of culture.

This fungus was isolated from soil at a depth of 45 centimeters (No. 13).

Actinomyces IV.

Colony circular, shiny, at first white, then dull cream, folds extending radially, forming tough leathery membrane on surface of culture.

This *Actinomyces* was obtained from pure sand of the beach of Lake Michigan (No. 10).

TABLE III

WORLD DISTRIBUTION OF GENERA ISOLATED IN ILLINOIS

Genus	England (7)	Germany (1)	Holland (18)	Japan (26)	Norway (9)	Iowa (2) (19)	Idaho (22)	Mich. (8) (20) (21)	N. J. (16) (33)	N. Y. (12)	R. I. (23)	Texas (35)
Alternaria	*		*	*		*	*	*	*	*	*	
Aspergillus	*	*	*	*		*	*	*	*	*	*	*
Chaetomium						*			*	*		
Circinella												
Cunninghamella						*		*		*		
Fusarium	*					*	*	*	*		*	*
Hormodendrum			*			*		*	*	*		
Mucor	*	*	*	*	*	*	*	*	*	*	*	*
Myrothecium									*			
Penicillium	*	*	*	*		*	*	*	*	*	*	*
Rhizopus	*	*		*	*	*	*	*	*	*	*	*
Stachybotrys						*				*		
Stysanus			*					*	*			
Trichoderma	*		*	*		*	*	*	*	*	*	
Trichosporium												
Trichurus												
Verticillium	*					*	*	*	*			
Zygorrhynchus	*			*		*			*	*	*	

The numbers given above refer to the bibliography.

DISCUSSION

In Table III is shown the occurrence in other parts of the world of the genera isolated in this investigation. A comparison of Tables II and III will show a striking agreement between the genera found most frequently in local soils and those reported most frequently from other localities. The writer found species of *Aspergillus*, *Mucor*, *Penicillium* and *Trichoderma* to be most numerous, with *Fusarium*, *Rhizopus* and *Zygorrhynchus* ranking next. Table III indicates that these fungi are those most often

reported from other parts of the world. This would seem to indicate that these genera are not merely transients, but fairly constant inhabitants of the soil throughout the world.

However, in any examination of soil from a new region, new forms may be expected. Abbott (**3**) in 1926 assembled a list of fungi which had been reported up to that date as occurring in the soil. After a careful review of the literature since Abbott's publication, particularly the work of Paine (**19**) in 1927, the following species from Illinois soils are found to be herein reported for the first time: **Chaetomium subterraneum,** *Circinella simplex*, *Mucor griseo-lilacinus*, *Myrothecium convexum*, *Stysanus medius*, *Trichosporium nigricans* Sacc. f. *lignicola*, **Trichurus terrophilus,** and *Verticillium lateritium*.

Whereas the writer realizes that this investigation is by no means comprehensive, it should serve as an introduction to the assembling of a fungous flora of local soils, as an incentive for further work, in which possibly fungous physiology as well as taxonomy may be considered, and also as a contribution to the slowly growing body of evidence for a definite and characteristic mycological flora of the soil.

SUMMARY

Isolations of fungi from twenty-two samples of Illinois soil have been made. The soils ranged in type from pure sand of the shore of Lake Michigan to humus of wooded areas. Samples were taken from cultivated and uncultivated areas, at the surface and at depths of 5, 15, 25, 45, 60, 90 and 120 centimeters.

Examination of pure cultures of the fungi yielded thirty-nine species and one variety, belonging to eighteen different genera. In addition, four *Actinomyces* and several yeasts were observed.

Fungi were most numerous at the surface of the soil, decreasing markedly with depth. A few forms, however, were found to be present at depths heretofore unexamined; namely, *Actinomyces* spp., *Penicillium* sp., *Trichoderma Koningi*, and *Zygorrhynchus Vuilleminii* at 60 centimeters; *Penicillium* sp., *Verticillium lateritium*, and *Zygorrhynchus Vuilleminii* at 90 centimeters; and **Chaetomium subterraneum,** *Penicillium* sp., and *Trichoderma Koningi* at 120 centimeters.

In tilled areas species of *Mucor* were dominant; in untilled areas they occurred only sparsely.

In surface soil species of *Mucor*, *Aspergillus* and *Penicillium* were most numerous; in sub-surface soil, *Penicillium* spp., *Trichoderma Koningi*, and *Zygorrhynchus Vuilleminii* appeared more frequently than other forms.

The following eight species are herein reported for the first time as occurring in the soil: **Chaetomium subterraneum** Swift & Povah, sp. nov., *Circinella simplex* van Tiegh., *Mucor griseo-lilacinus* Povah, *Myrothecium convexum* Berk., *Stysanus medius* Sacc., *Trichosporium nigricans* Sacc. f. *lignicola*, **Trichurus terrophilus** Swift & Povah, sp. nov., and *Verticillium lateritium* Berk.

Chaetomium subterraneum and **Trichurus terrophilus** are described as new species.

The conclusions of previous workers with regard to the genera found most commonly in the soil: namely, *Aspergillus*, *Fusarium*, *Mucor*, *Penicillium*, *Rhizopus*, *Trichoderma*, and *Zygorrhynchus*, are confirmed by the present work.

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EXPLANATION OF PLATES

Measurements based on original drawings which were reduced about one-third in reproduction

PLATE 16

Trichosporium nigricans Sacc. f. *lignicola*

Fig. 1. Swollen mycelium, showing oil globules and chlamydospores. (× 1000.)

Fig. 2. Septate mycelium, bearing globular, guttulate conidia on short conidiophores. (× 1000.)

Circinella simplex van Tiegh.

Fig. 3. Habit sketch of conidiophore, showing manner of branching. (× 20.)

Fig. 4. Detail of sporangiophore. *A*. Columella (× 375) and spherical spores (× 750). *B*. Spherical sporangium. (× 375.)

Verticillium lateritium Berk.

Fig. 5. Detail of verticillately branched conidiophore. (× 1000.)

Fig. 6. Conidia. (× 1000.)

Penicillium roseum Link (?)

Fig. 7. Conidiophore, showing secondary branches and attached conidia. (× 1000.)

Fig. 8. Habit sketch of branching conidiophore, arising from rope-like mass of hyphae. (× 300.)

PLATE 17

Aspergillus luchuensis Inui

Fig. 1. Echinulate spores. (× 2000.)

Fig. 2. Vesicle, showing arrangement of sterigmata and spores. (× 500.)

Fig. 3. Swollen mycelium with chlamydospores. (× 500.)

Aspergillus fumigatus Fres.

Fig. 4. Vesicle with attached sterigmata. (× 400.)

Fig. 5. Columnar spore bodies. (× 150.)

Fig. 6. Portion of conidial chain. (× 1000.)

Myrothecium convexum Berk. & Curt.

Fig. 7. Portion of vegetative mycelium. (× 1000.)

Fig. 8. Sporodochia, each with margin of sinuous cilia. (× 150.)

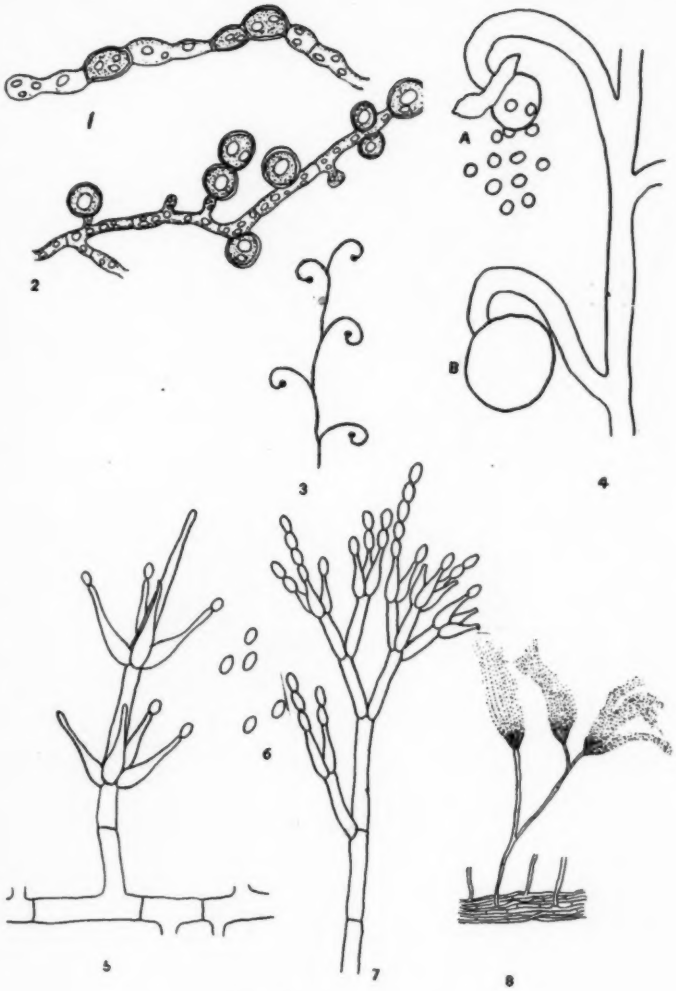
Fig. 9. Warty, branched conidiophore. (× 1000.)

Fig. 10. Fusiform spores. (× 1000.)

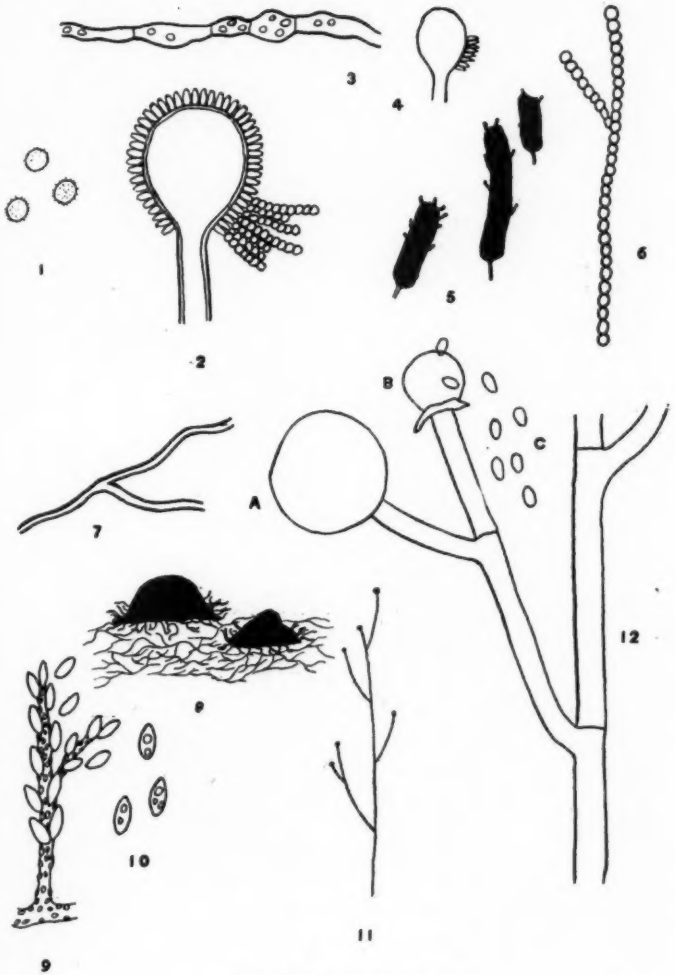
Mucor griseo-lilacinus Povah

Fig. 11. Habit sketch of conidiophore, showing manner of branching. (× 20.)

Fig. 12. Detail of sporangiophore. *A*. Sporangium. (× 375.) *B*. Columella with basal collar. (× 375.) *C*. Spores. (× 800.)



SOIL FUNGI OF ILLINOIS



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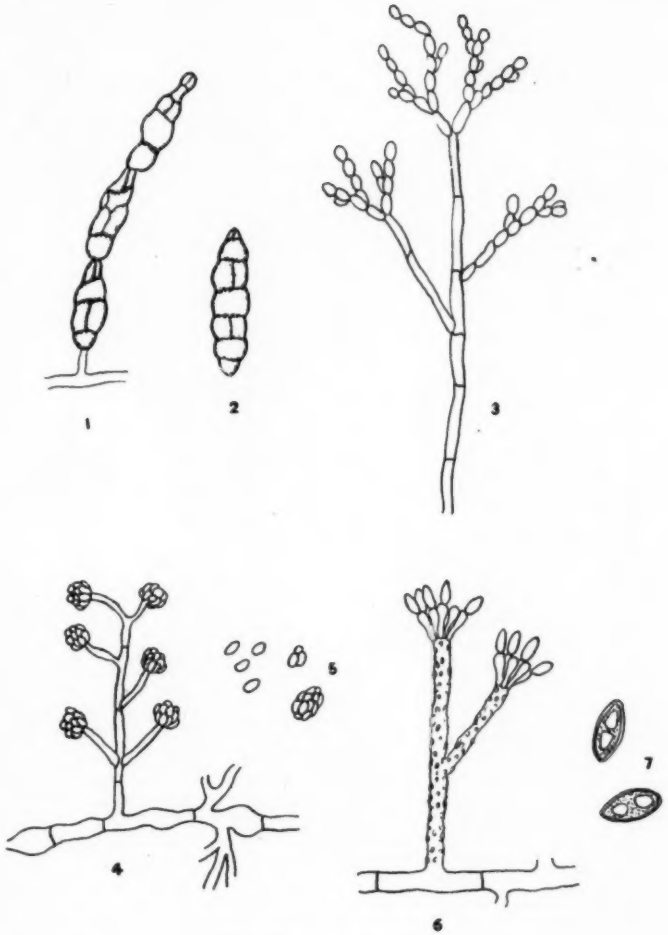

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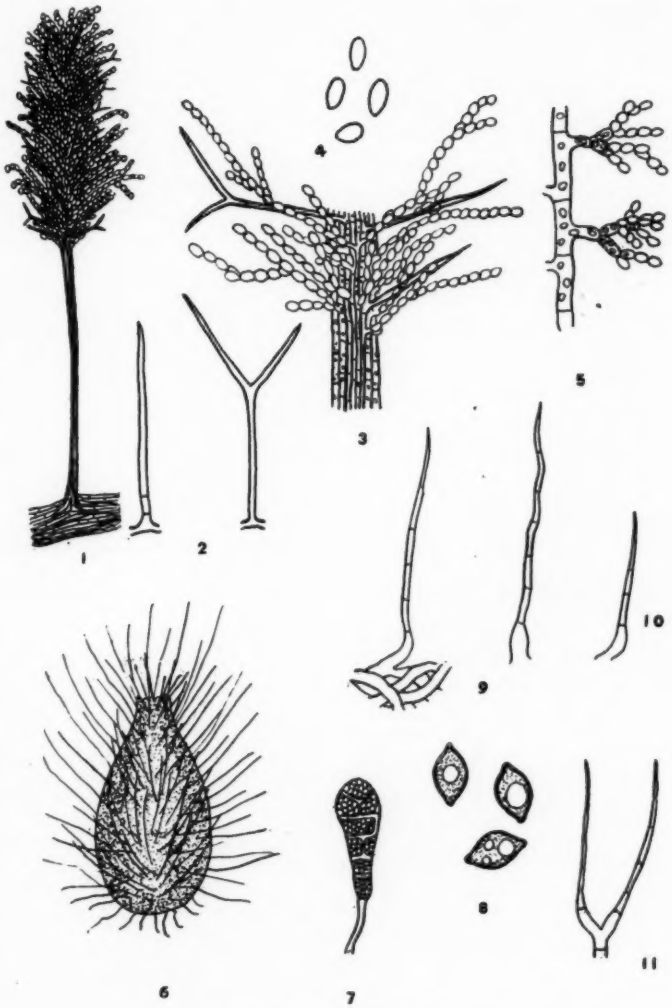

SOIL FUNGI OF ILLINOIS

PLATE 18

Alternaria humicola Oud.

Fig. 1. Chain of spores on short conidiophore. (× 1000.)
Fig. 2. Single muriform spore. (× 1000.)

Hormodendrum cladosporioides Fres.

Fig. 3. Detail of conidiophore, showing manner of branching, with attached catenulate conidia. (× 1000.)

Trichoderma Koningi Oud.

Fig. 4. Branched conidiophore, bearing spherical conidial heads. (×1000.)
Fig. 5. Detached conidia, single and in groups. (× 1000.)

Stachybotrys atra Corda

Fig. 6. Warty, branched condiophore, showing whorl of sterigmata with attached spores. (× 1000.)
Fig. 7. Fusiform spores with large oil globules. (× 1500.)

PLATE 19

Trichurus terrophilus Swift & Povah, sp. nov.

Fig. 1. Conidial body rising from rope-like aggregation of mycelium, showing slender stalk and fertile portion bearing catenulate conidia and simple and forked setae. (× 250.)
Fig. 2. Details of simple and forked setae. (× 1000.)
Fig. 3. Lower portion of fertile part of fruit body, showing chains of spores emerging laterally from closely associated hyphae. (× 500.)
Fig. 4. Spores. (× 2000.)
Fig. 5. Young hypha, showing early stage in spore development. (×1000.)

Chaetomium subterraneum Swift & Povah, sp. nov.

Fig. 6. Ovoid perithecium, showing uniform distribution of simple, straight, lateral and terminal hairs, with shorter setae immediately surrounding ostiole. (× 250.)
Fig. 7. Immature ascus containing young spores. (× 500.)
Fig. 8. Lemon-shaped spores. (× 1500.)
Fig. 9. Straight seta, showing interlacing hyphae of perithecial wall, and slightly undulate seta with bulbous swelling at base. (× 500.)
Fig. 10. Short seta from ostiole region of perithecium, showing bulbous base and elbow-turn. (× 500.)
Fig. 11. One of the occasional branched setae from lower portion of perithecium. (× 500.)

THE NATURE OF GIANT SPORES AND SEGREGATION OF SEX FACTORS IN NEUROSPORA

B. O. Dodge

The occasional production of unisexual spores in asci of *Neurospora tetrasperma*, which is normally four-spored but occasionally may produce five or six spores in a single ascus, is fortunate, as it enables one to cross this homothallic species with the heterothallic species of the genus. Unisexual haplonts of *N. tetrasperma* may be secured in two different ways. First,

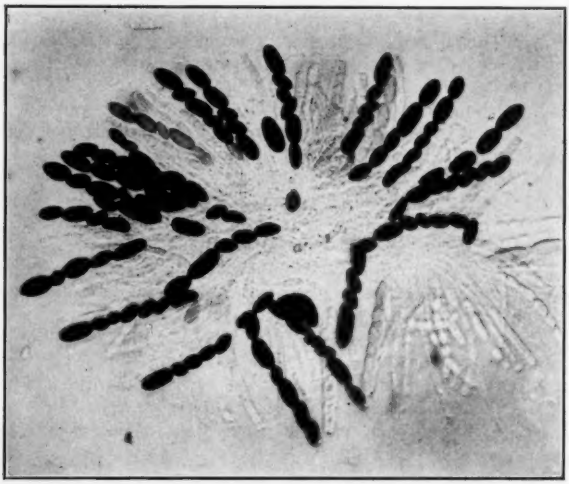

Fig. 1. Asci from a hybrid perithecium obtained by back-crossing an f_1 hybrid (*Neurospora sitophila* × *N. tetrasperma*) with the *tetrasperma* parent. No abortion of ascospores apparent. Large ascospores probably contain several nuclei. × 190.

one can select the very small ascospores which give rise to unisexual mycelia. Such haplonts can be propagated indefinitely asexually either by transplanting cuttings of their mycelia or by

sowing their conidia which are, after all, for purposes of propagation, nothing but small cuttings. Unisexual haplonts may also be secured if the conidia from normal bisexual mycelia are plated out and selected on the basis of the length of time required for their germination. Choosing those conidia whose germination was the longest delayed, one obtains 10 to 25% unisexual mycelia. Regardless of which way the unisexual haplonts are obtained normal perithecia will develop when haplonts of opposite sex are mated in culture. The ascospores will be predominantly bisexual as usual.

Certain back-crosses in the series started by crossing *N. sitophila* and *N. tetrasperma* tend to produce one or two very large spores in some of the asci.[1] One or more smaller spores may develop along with a single giant spore (FIG. 1). The largest spores may have as many as four nuclei of each sex when they are cut out. The ascus then would be 1-spored. The inclusion of more than one nucleus in an ascospore at its delimitation is characteristic of the *N. tetrasperma* parent. Over-sized spores are formed occasionally by the parent species, *N. tetrasperma* (FIG. 2, x), but in such cases the big spores are the same genotypically as any other spore except that the very small spores are unisexual.

In quite another category genetically are the giant spores of hybrids. The fact that some asci in a back-cross produce giant spores while some other ascus in the same perithecium may contain eight uninucleate spores, is an expression of *tetrasperma* as contrasted with *sitophila* inheritance. The ascus being a mother cell, whatever segregations occur here would be represented by different types of nuclei which result from the three successive nuclear divisions. When all eight nuclei happen to be included in a single giant spore which can be grown into a new plant, the complexity of its composition genetically is such as to require careful analysis if its total inheritance is to be ascertained. With each mycelial cell containing several nuclei which may differ genotypically there is not likely to be an equal distribution of all of the elements of inheritance to different hyphal branches.

[1] Dodge, B. O., The Production of Fertile Hybrids in the Ascomycete *Neurospora*, Jour. Agr. Research, **36**: 1–14, 1928.

Since the ascogenous cells are also multinucleate there are bound to be all sorts of matings of nuclei in the various asci of the same fruit body.

Why breed from a complex giant spore when it is simpler to start with the small uninucleate spores? These back-cross asci rarely develop eight spores, and even when they do there are the hazards to be met in their separation and germination. Furthermore a study of the progeny of the eight separate spores in an ascus would not tell us anything about the peculiarities of the giant spores as such.

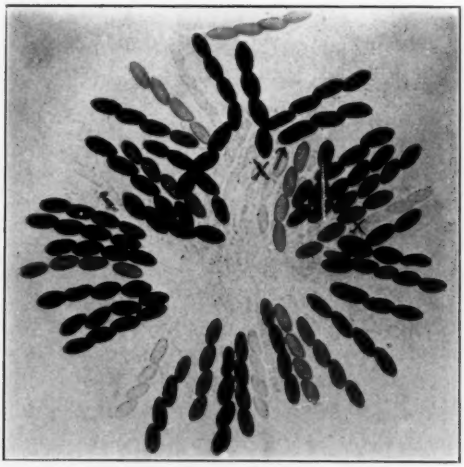

FIG. 2. Asci from a perithecium of *Neurospora tetrasperma*. At "x" asci with three spores, one spore over-sized in each case. × 190

There is a way out of the difficulty. The method is equivalent to cutting up the giant spore into its components and then growing each part independently in pure culture. The inheritance carried by each nucleus could thus be studied by itself and in any combination desired. The procedure would simply be to grow a mycelium from the giant hybrid ascospore and plate out the uninucleate unisexual conidia in the way noted previously.[2] Just how many distinct types of haplonts could be

[2] Dodge, B. O., Unisexual Conidia from Bisexual Mycelia, MYCOLOGIA, **20**: 226–234, 1928.

isolated would depend on the nature of the sexual processes involved in the production of the ascocarp. Of course there would be two kinds of mycelia as regards their sex. Provided the factors are not sex-linked we would get also different types of haplonts with regard to other characters, the number depending on just what had been the nuclear behavior previously in the life cycle. Perhaps we have here something which will eventually throw light on the question as to the nature of sexual reproduction in the ascomycetes as distinct from nuclear fusions in the ascus. Stout [3] has recently discussed the clon in the fungi in his treatment of clons in the higher plants.

The mycelium obtained by germinating a giant hybrid ascospore would also produce a great many multinucleate conidia. The type of clon obtained from a multinucleate conidium must therefore depend on which nuclei happen to have been thrown together in that conidium. There could, therefore, be selected out just as many different types of clons as there would be possible combinations of the genotypically different nuclei contained within the original ascospore. The complications met with in connection with giant hybrid ascospores would not enter into a study of crosses between *N. sitophila* and *N. crassa* because in both species the ascospores are normally uninucleate and unisexual at their origin.

This brings us up to the point where the rare or occasional giant spore is developed in asci of the heterothallic species, *N. crassa*. It has been assumed that since the large spores of *N. tetrasperma* are bisexual while the little ones are unisexual, the same line of reasoning should hold for the abnormal spores of *N. crassa*. Its normal ascospores, which are about 31 μ long, are unisexual. Its giant spores, 60 to 90 μ long, should then be bisexual and give rise to homothallic instead of unisexual haplonts. The determination of the sexual nature of these giant ascospores by culture work shows that this line of reasoning does not hold. Moreover it has been found that segregation of the sex factors in *N. crassa* must take place in the first nuclear division in the ascus. The distribution of the spores as regards their

[3] Stout, A. B., The Clon in Plant Life, Jour. N. Y. Bot. Gard., **30**: 25–37, 1929.

sex does not agree with the report by Marguerite Wilcox [4] for *N. sitophila*, a species in many respects very similar to *N. crassa*.

GIANT SPORES OF *Neurospora crassa*

As noted previously one finds occasionally a very large ascospore in a spore print of *N. crassa*. A crushed mount may also show a 4-spored ascus (FIG. 3, RIGHT). These 4-spored asci are such perfect pictures of normal asci of *N. tetrasperma* (FIG. 2) that one would expect the spores would also be bisexual. Hoping

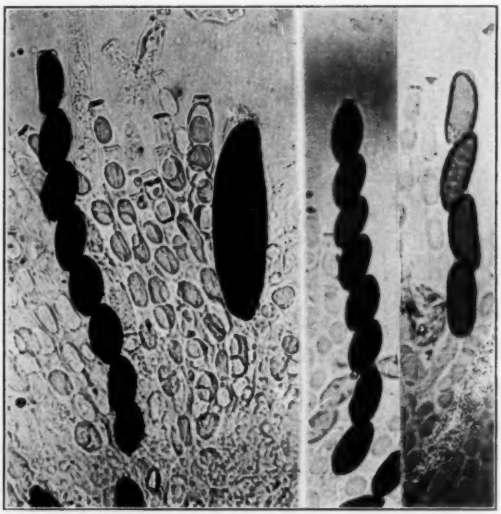

FIG. 3. Asci from a perithecium of *Neurospora crassa*. At the center a giant spore and to the left a normal ascus for comparison. The two asci at the right are from the same perithecium. The 4-spored ascus resembles a normal ascus of *N. tetrasperma*. × 300.

to obtain a homothallic mycelium from a giant spore such as is shown in figure 3, several large spores were isolated and germinated. Cultures were obtained from spores 41, 49, 50, 51, 58, 64 and 85 microns in length. After the germ tubes from each end of a spore had branched out sufficiently the tip ends were cut off

[4] Wilcox, M. S., The Sexuality and Arrangement of the Spores in the Ascus of *Neurospora sitophila*, MYCOLOGIA, **20**: 3–16, 1928.

and transplanted. Several cultures from each spore were obtained in this way. This method was used to avoid possible contamination from conidia which must occasionally be carried over with the ascospore.

It was certainly expected that the mycelium from the largest spores would prove to be homothallic and develop perithecia. Such was not the case, as all the cultures remained sterile. On the theory that the nuclei of opposite sex might have been distributed to different hyphal branches, all of the mycelia derived from the same ascospore were grown together in pairs in all possible combinations. For example, 13 separate cultures had been obtained from the largest spore, which was 85 μ long. None of the combination cultures produced perithecia. It is clear that the mycelia obtained from these particular spores at least were all unisexual. This suggested that perhaps the segregation of the sex factors in *N. crassa* might occur during the first of the three nuclear divisions in the ascus, so that the four nuclei in one end would all be alike sexually. Unless a giant spore in one end of the ascus included more than four nuclei at its origin it would be unisexual.

Culture work to determine just where segregation takes place was begun by germinating normal ascospores. Meanwhile further study of crushed mounts showed that in *N. crassa* oversized spores are very frequently present in asci showing abortion of other spores. In this case a giant spore may include only one nucleus at its origin but grow disproportionately because of food made available by abortion of adjacent spores. But such 4-spored asci as are shown in figure 3 do not present evidence of spore abortion. If the spindles of the third division in the ascus are oriented as they are in *N. sitophila*,[4] each of the four spores might include two sister nuclei. This would account for their size and unisexual nature. There is still the possibility of nuclear degeneration without spore formation as described by Harper.[5] This point was discussed briefly by the writer in another connection.[6] A cytological study of the ascus of *N. crassa* will be neces-

[5] Harper, R. A., Kerntheilung und freie Zellbildung im Ascus, Jahrb. Wiss. Bot., **30**: 249–284, 1897.

[6] Dodge, B. O., Formation of Spores in Asci with Fewer than Eight Spores, MYCOLOGIA, **22**: 8–21, 1928.

sary to determine the question of nuclear degeneration raised here.

SEGREGATION OF THE SEX FACTORS IN 8-SPORED ASCI IN *N. crassa*

Mature ascospores from several normal asci were isolated one by one and their positions in the ascus were noted. After germination each spore was transferred directly to a tube of corn meal agar. As no perithecia developed in these single ascospore cultures it was clear that no conidia of opposite sex had been carried over in any case with the transfer of the ascospore. Cultures were obtained in this way from all eight spores in each of three different asci. In each case the eight haplonts were grown together in pairs in all possible combinations. The results obtained by growing the three sets of haplonts as noted all agreed. The checkerboard diagram of these results is shown in Table 1.

TABLE 1

RESULTS OBTAINED BY GROWING IN ALL POSSIBLE COMBINATIONS EIGHT HAPLONTS REPRESENTING THE EIGHT ASCOSPORES FROM AN ASCUS OF *N. crassa*

Spores are numbered in order of their position in the ascus. The sign + indicates that perithecia were produced; the sign − negative results.

	1	2	3	4	5	6	7	8
1	−	−	−	−	+	+	+	+
2	−	−	−	−	+	+	+	+
3	−	−	−	−	+	+	+	+
4	−	−	−	−	+	+	+	+
5	+	+	+	+	−	−	−	−
6	+	+	+	+	−	−	−	−
7	+	+	+	+	−	−	−	−
8	+	+	+	+	−	−	−	−

The table shows that no perithecia were produced in culture when haplonts nos. 1, 2, 3 and 4 were grown separately or together in any combination; the same is true for haplonts nos. 5, 6, 7 and 8. But when any one of the first four haplonts is grown with any one of the second four, perithecia are produced. This proves

conclusively that the four spores in one end of the ascus were of the same sex.

The results were further checked by isolating the spores from three other asci. But here no attempt was made to record the exact position of the spore except that in each case the four spores from one end of the ascus were first carefully isolated from the four in the other end, after which the spores in each group were separated and germinated. In one case only 3 spores from each end of the ascus germinated. This was sufficient for the purpose however. If segregation had occurred in the second division, one of the three spores would differ in sex from the other two. Either three or four spores from one end of each of ten other asci were also isolated and grown in culture. In each of the thirteen sets noted the mycelia were grown in pairs in various combinations and in addition the mycelia representing the four spores in the one end of the ascus were all grown together in one culture. This last procedure, after all, furnishes the very best of evidence that all four spores are alike sexually. If they were not alike, perithecia would surely have developed. The results of the work represented by several hundred cultures go to prove conclusively that in *N. crassa* segregation of the sex factors occurs in the first nuclear division in the ascus.

SEGREGATION OF SEX FACTORS IN *N. sitophila*

Wilcox, working with *N. sitophila*, concludes that segregation of the sex factors in that species must occur in the second nuclear division in the ascus. She reaches this conclusion on the basis of the orientation of the spindle figures presented during the three divisions, taken in connection with the results of her culture work. She reports, as noted previously, that the spores alternate in pairs in the ascus, two of one sex and two of the other.

Judging from what is said to occur in species of *Coprinus* it would not be strange to find two species of the same genus of ascomycetes differing with respect to the particular nuclear division during which segregation takes place. The asci in *N. crassa* and *N. sitophila* are much alike, being long and slender, and their eight spores are regularly uniseriate. No doubt the spindles of the three divisions have much the same orientation. The ques-

tion arises as to whether they really differ so markedly as to the disposition of their spores of opposite sex.

The eight spores in an ascus of *N. sitophila* were carefully isolated and the positions which they had occupied in the ascus were noted. The spores were germinated and two complete sets of cultures in duplicate were obtained. After three or four days the cultures in both sets showed striking differences in the color and quantity of conidia produced. Cultures nos. 1, 2 and 5, 6 showed very pale fluffy aerial hyphae with no conidia. Cultures nos. 3, 4 and 7, 8 showed masses of bright orange-colored conidia. Clearly here was an alternation in pairs which suggested that the mycelia would also alternate in pairs as to their sex. The eight haplonts were then grown together in pairs in all possible combinations. As a further check haplonts nos. 1, 2, 3 and 4 were all grown together in one culture, as were haplonts nos. 5, 6, 7 and 8. The results show clearly that, whatever may be the meaning of the definite alternation in pairs viewed from the standpoint of production and color of conidia, it has no significance as regards the sex of the ascospores in this ascus. Spores nos. 1, 2, 3 and 4 proved to be all alike and opposite in sex to spores nos. 5, 6, 7 and 8. A checkerboard diagram showing results of the culture work would be the same as that given for *N. crassa* in Table 1.

It is then interesting to prove that segregation of the factors determining the type of conidia produced by a mycelium of *N. sitophila* occurs in the second division in the ascus, while the factors for sex are segregated in the first division. Cultures made to determine whether distinct races can be obtained by mating haplonts nos. 1 and 5 as contrasted with the results obtained by mating haplonts nos. 3 and 7 give very positive and striking results.

Further work proves that in *N. sitophila* the spores may sometimes also alternate in pairs as to their sex as reported by Wilcox. In the case of *N. tetrasperma* each of the four spores in the ascus is bisexual because it includes at its origin one nucleus of each sex. Ascospores of *N. crassa* and *N. sitophila* are unisexual and only a single nucleus is included in a spore when it is cut out. In the first species the four spores in one end of an ascus are all of the same sex and they are of the opposite sex to

the four spores in the other end of the same ascus. In *N. sitophila* the segregation of the sex factors may occur in either the first or second division. A full account of the work on developing new strains of *N. sitophila* which are practically sterile so far as conidia are concerned will be published later.

THE NEW YORK BOTANICAL GARDEN

NOTES AND BRIEF ARTICLES

In my article in MYCOLOGIA, March–April number, 1929, page 98, "A" under legend for Figure 1 appears "Ascospores," which should read "Basidiospores."—S. M. ZELLER.

Professor A. H. R. Buller of the University of Winnipeg, Canada, an Associate Editor of MYCOLOGIA, has recently been elected Fellow in the Royal Society of London.

The New York Botanical Garden has recently added to its already large rust collection about eight hundred specimens of GERMAN UREDINEAE distributed by Theodor Oswald Weigel of Germany.

The Editor of this publication is planning to spend two months during the summer in the Rocky Mountains near Denver continuing his field studies on the Cup-fungi preparatory to publishing a volume on the inoperculate forms which will be a companion volume to the one just issued on the Operculates. So much interest has been manifest in this work that the project is being pushed with renewed interest.

The Herbarium and Library of the late Dr. Bruce Fink has been acquired by the University Herbarium of the University of Michigan. His collection of Lichens especially has great scientific value because of its being the source material on which the forthcoming book of Dr. Fink's "Lichens of the United States" has been based. The material will be transferred from Miami University in June and will be accessible to scientists within a few months thereafter.—C. H. KAUFFMAN.

THE IMPERFECT STAGE OF *Crytosphaeria populina* [1]

Perithecia of *Crytosphaeria populina* (Pers.) Sacc. growing on a dead branch of the Balm of Gilead Poplar, were brought into the laboratory during Oct. 1928. Pieces of the dead bark containing perithecia were laid on wet blotting paper in a petri dish. A small amount of nutrient agar was poured on the inside of the petri dish cover, and when this had solidified the cover was placed on the dish with the agar about one cm. above the bark.

In a few days the allantoid, one-celled ascospores were found in dense masses on the agar. They germinated immediately and several transfers were made from the germinated spore masses to nutrient agar in culture tubes. While these were not single spore cultures there appeared to be no contaminations in the petri dishes at the time the transfers were made.

Within from two to four weeks pycnidia were formed in the tube cultures. Orange-yellow spore masses were pushed out of these pycnidia, in some cases forming tendrils entirely similar to those formed by species of *Cytospora*.

Examination of the spores showed that they belong to the genus *Cytosporina*. The spores were hyaline, filiform, sickle-shaped, some varying to hook-shaped, and averaged 20 $\mu \times 1\ \mu$. The length was measured across the longest axis of curvature of the spore, and varied from 15 μ to 30 μ, depending on the degree of curvature.

After these results had been obtained, the writer found that Wehmeyer had also grown *Cryptosphaeria populina* in culture. In his paper in the American Journal of Botany (**13**: 1926), in a footnote at the bottom of page 592, he mentions the occurrence of pycnidia with spores such as described above, in cultures of *Crytosphaeria populina* on sterilized poplar twigs.

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[1] Abstract of a report presented to the Conference of the Scientific Staff and Registered Students of the New York Botanical Garden on March 13, 1928.